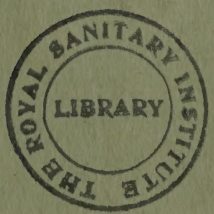
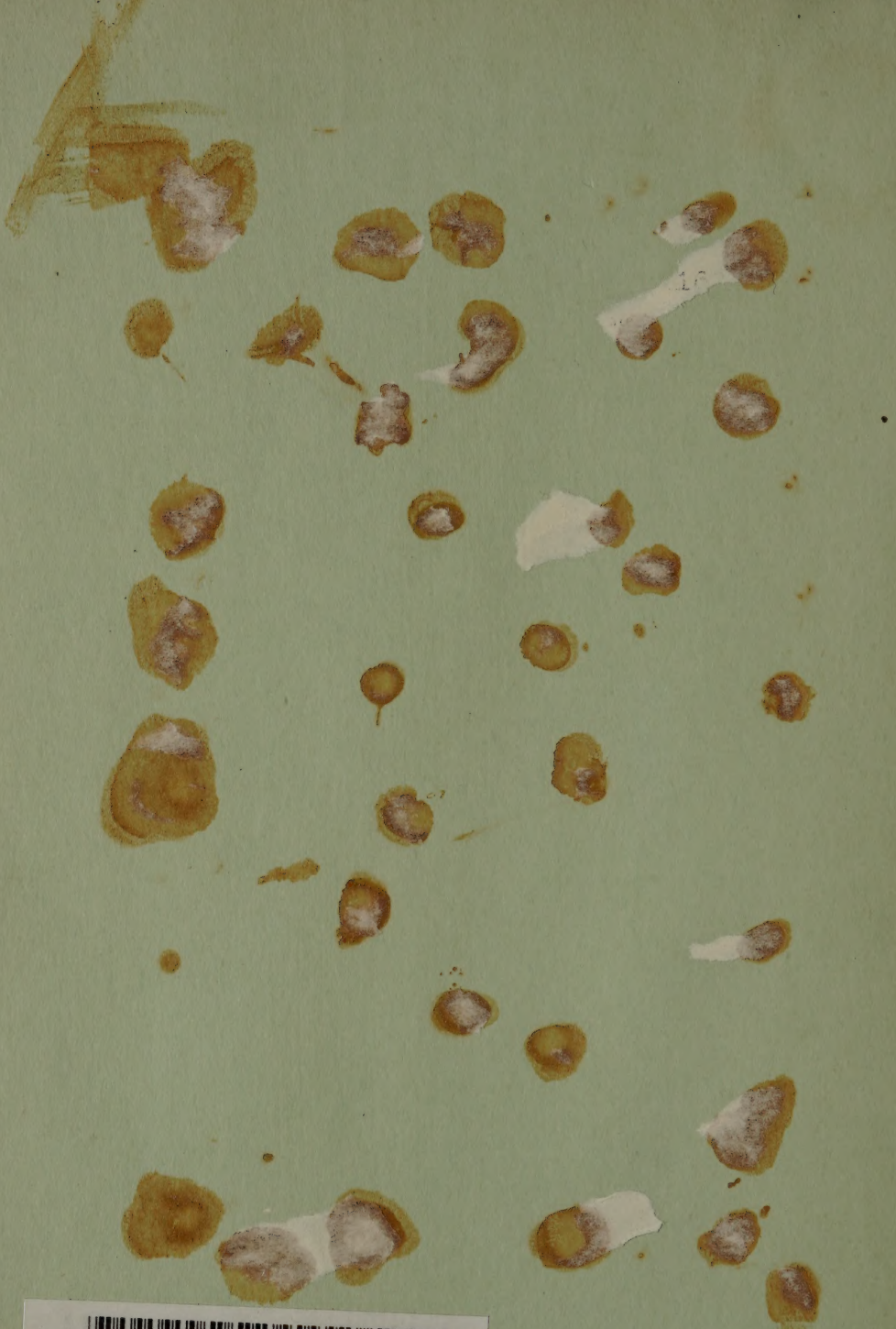
PHYSICIANS' GUIDEBOOK TO PUBLIC HEALTH LABORATORY SERVICES

Second Edition

1954



Connecticut State Department of Health
Stanley H. Osborn, M.D., Dr.P.H., Commissioner
Hartford, Connecticut



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A laboratory test is no better than the specimen and the specimen no better than the manner in which it was collected

Connecticut State Department of Health
Stanley H. Osborn, M.D., Dr.P.H., Commissioner
Hartford, Connecticut

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PREFACE

The purpose and form of this volume have been described in the preface to the first edition (1945) which is reproduced below. In the years since the first edition was published, laboratory science has progressed and the addition of new tests of value in the diagnosis of disease has made a revision necessary. The present volume has benefited from constructive criticism of the first edition. To all those who have so kindly written their suggestions and comments for improvement of this "Guidebook," the Connecticut State Department of Health extends its grateful appreciation. Once again we acknowledge a special debt of gratitude to the American Public Health Association for graciously giving permission to use data compiled in their official report "The Control of Communicable Diseases in Man," 7th Ed., 1950.*

PREFACE TO FIRST EDITION

For some time there has been a need for a publication that might be placed in the hands of every physician in Connecticut to furnish in "ready-reference" form information regarding the many diagnostic services that are now available at the Bureau of Laboratories of the Connecticut State Department of Health. This "Physicians' Guidebook to Public Health Laboratory Services" has been issued to meet that need. The volume is a contribution of the administrative staff of the Bureau of Laboratories. It has in no sense been hastily prepared but rests on a foundation of many years of laboratory experience buttressed by an appreciation of the many and intricate diagnostic problems that present themselves in the field of communicable diseases — a coign of vantage that can be attained only through frequent interchange of ideas with physicians, epidemiologists and health officers. The extent to which this little book is found useful will be the real test of its value to the physicians of the State.

The primary purpose of the undertaking has been to furnish information for each communicable disease under the headings of "Current laboratory services" that are available to physicians; "Collection of specimens" that are indicated worthwhile; and "Limitations of laboratory tests" that may be performed upon request. While we were engaged in the preparation of the manuscript, the Sixth Edition of "The Control of Communicable Diseases" was published by the American Public Health Association and it seemed to us that certain additional information from that volume would add greatly to the usefulness of this manual. Through permission kindly granted by Reginald M.

*"The Control of Communicable Diseases, An Official Report of the American Public Health Association" (7th Ed., 1950, published by the American Public Health Association, 1790 Broadway, New York, N.Y., Price of single copies 40 cents) contains additional information for each of the communicable diseases of this comprehensive list under the headings: "Recognition of the disease," "Incubation period," "Period of communicability," "Susceptibility and immunity," and "Methods of control." That publication is suggested as a worthwhile companion to this "Guidebook" on the desk of every physician in Connecticut.

Atwater, M.D., Executive Secretary of the American Public Health Association, it has been possible to include verbatim in the "Guidebook" the additional authoritative statements for each disease that appears under the headings: "Etiologic agent," "Source of infection," "Mode of transmission," and "Prevalence."

The puzzling question of which diseases to include in this compendium and which to omit was solved through a decision to include all of the diseases listed under the 72 sections in the "Control of Communicable Diseases." The reason for doing this is the seemingly sound one that the selection for that volume has resulted from a 20-year period of study by a carefully selected continuing committee of prominent authorities in the administrative control of communicable diseases. With the adoption of the list there are included in this volume some diseases that are rarely if ever seen in Connecticut; however, it would be presumptuous to say with respect to almost any disease included that it will not appear in our midst under post-war conditions, at least as an isolated case. Under these circumstances we have preferred to err by including a disease that may never appear in this State rather than to omit data that may be of real worth to a physician in an emergency when the unexpected occurs. In addition, ten groups of diseases* have been added which appeared of importance from the standpoint of this publication.

The Connecticut State Department of Health offers this manual to the physicians of the State in the belief that it is as complete as is desirable to answer the questions that will ordinarily arise in regard to the laboratory tests currently available for public health purposes; as concise as such work can be; as authoritative and up-to-date as any reference book available anywhere for the purpose. Comment and criticism will be welcomed. It is hoped that physicians generally will find the "Guidebook" useful enough so that instead of becoming a dust covered volume that is seldom or never used it will be a frequently consulted reference on the desk of many a physician.

FRIEND LEE MICKLE, SC.D.

OUTFITS FOR COLLECTION OF SPECIMENS

Specimen outfits are referred to in the text by symbols. These outfits may be obtained from the Bureau of Laboratories upon request. When ordering, use symbols.

*The ten sections on "Cestode Infections, miscellaneous," "Gas Gangrene," "Haverhill Fever," "Hydatid Disease," "Mycotic Infections, miscellaneous," "Pinworm Infection," "Protozoan Diseases, miscellaneous," "Trematode Infections," "Trichuris Infection," and "Vincent's Infection (Trench mouth), Fusospirochetosis" appear only in the "Guidebook" and were not taken from publication cited.

ACTINOMYCOSIS

1. *Etiologic agent.* *Actinomyces bovis* and other species of this genus.

2. *Source of infection.* Unknown. Possibly infected animals or contaminated grasses, and possibly extension of a previously existing saphrophytic infection in the oral cavity.

3. *Mode of transmission.* Among cattle, principally by grains, grasses, cattle fodder, and stable bedding contaminated by discharges from lesions of the disease, infected abrasions or wounds of oral cavity or body surface. It is not probable that the disease is transmitted from man to man. It may be transmitted from animal to man, but only rarely and indirectly through infection of oral or skin wounds by contaminated materials. The disease sometimes follows extraction of carious or broken teeth, or accidental injury, particularly to the jaws. Veterinary instruments such as needles, scalpels and other equipment used by farmers when they are vaccinating or treating animals for local conditions may be the cause of infection.

4. *Prevalence.* Infrequent among humans.

5. *Current laboratory services.*

   A. Microscopic examination of direct and stained smears from abscesses or other lesions for typical "sulphur granules" and for *Actinomyces.*

   B. Cultures of material from abscesses or other lesions for *Actinomyces.*

6. *Collection of specimens.* Use "FI" outfit; if "sulphur granules" appear to be present, these are the best specimens; collect material from lesion and inoculate surface of slanted medium in bottle; also make smears on microscope slides in outfit.

7. *Limitations of laboratory tests.* Presumptive evidence of infection by microscopic examination should be confirmed by culture whenever possible.

AMEBIASIS; AMEBIC DYSENTERY

1. *Etiologic agent.* *Endameba histolytica.*

2. *Source of infection.* Feces of infected persons, especially carriers. Acute cases of little menace due to fragility of trophozoites.

3. *Mode of transmission.* Contaminated foods, especially those commonly served cold and moist; hand-to-mouth transfer of infected material from moist objects soiled with discharges of an infected individual; contaminated water; flies; direct transmission unusual.

4. *Prevalence.* In continental North America amebic dysentery is not common but amebiasis with other manifestations is being rec-

ognized with increasing frequency. Epidemic outbreaks are rare. It is estimated that almost 10 per cent of the population are carriers presenting no symptoms. More prevalent in the tropics and the Orient.

5. *Current laboratory services.*

   A. Microscopic examination for trophozoites and cysts of *Endameba histolytica.*

   B. Serologic test on blood serum.

6. *Collection of specimens.*

   A. Take feces from different parts of the stool and place portions in each of the bottles in "PD" outfit. Fill the bottle containing liquid to the shoulder; the bottle without liquid should be filled at least one-third full. In acute infections a liquid stool following a saline cathartic is recommended.

   B. Blood (10 ml.+) for complement fixation test may be submitted in "MI" outfit. Mark history blank plainly "For Amebiasis".

7. *Limitations of laboratory tests.*

   A. A single report showing no amebae in a stool is of little value for the exclusion of amebiasis; a series of specimens should be submitted. *Endameba histolytica* is generally regarded as the only pathogenic ameba and is the cause of amebic dysentery and its sequelae. The presence of *E. histolytica* cysts does not mean the presence of an active infection; some authorities have expressed the opinion that the "small race" of this species is non-pathogenic. Other amebae, such as *Endameba coli, Endolimax nana* and species of *Iodameba,* are non-pathogenic.

   B. The complement fixation test for amebiasis is useful when amebae cannot be demonstrated in stools, particularly when amebic abscess is suspected. Failure to obtain a reaction does not exclude amebiasis.

ANCYLOSTOMIASIS (HOOKWORM DISEASE)

1. *Etiologic agent. Necator americanus and Ancylostoma duodenale.* Infective larvae of cat and dog hookworms *Ancylostoma brasiliense* and *A. caninum* may cause a dermatitis called creeping eruption. These larvae are destroyed in the skin and do not otherwise affect man.

2. *Source of infection.* Usually soil contaminated with infective larvae from ova in stools deposited by infected persons.

3. *Mode of transmission.* The infective or third-stage larvae penetrate the skin, usually of the foot, and pass via the lymphatics to the inferior *vena cava* and the right heart, hence in the blood

stream to the lungs, where they pierce the capillary walls and pass into the alveoli. They then pass up the bronchi and trachea to the throat, whence they are swallowed and finally reach the small intestine, where they develop to maturity. The chief mode of infection is through the skin.

4. *Prevalence.* Widely endemic in areas having favorable soil, moisture and temperature for development of infective larvae.

5. *Current laboratory services.* Microscopic examination of feces for ova (or larvae) of *Necator americanus* and *Ancylostoma duodenale.*

6. *Collection of specimens.* Use "PD" outfit. With a tongue depressor or similar implement take specimens of feces from different parts of the stool shortly after it is passed and place in bottles supplied with outfit.

7. *Limitation of laboratory tests.* Since hookworms do not multiply in the body, a light infection may not mean clinical hookworm disease. For the evaluation of treatment a series of negative stool examinations has more value than a single specimen.

ANTHRAX

1. *Etiologic Agent.* Anthrax bacillus, *Bacillus anthracis.*

2. *Source of infection.* Hair, wool and hides and their manufactured products, flesh, blood and feces of infected animals, and manufactured products such as bone meal. Cattle, sheep and horses are the animal hosts of chief importance although many species may be infected.

3. *Mode of transmission.* Inoculation as by wound or scratch, inhalation of spores, ingestion of insufficiently cooked meat, and mechanically by flies.

4. *Prevalence.* Rare and sporadic in humans and associated only with the occurrence of the disease in animals, or with handling hide and hair or other products from infected animals. Local epizootics occur in cattle and sheep.

5. *Current laboratory services.* Isolation and identification of *Bacillus anthracis* by culture and animal inoculation of body fluids and of suspected sources of infection derived from animals.

6. *Collection of specimens.* Collect pus, exudate or other body fluid on sterile swab contained in "HS" outfit. Follow directions on history blank in "HS" outfit but mark history blank plainly "For anthrax". In suspected pneumonic infections submit sputum in sterile bottle in "MI" outfit.
Before submitting specimens from suspected sources of infection contact Bureau of Laboratories.

7. *Limitations of laboratory tests.* Suspicious cultural findings must be confirmed by animal inoculation tests. This procedure is nec-

essary to distinguish between *Bacillus anthracis* and common saprophytic spore-forming organisms. Since anthrax bacilli are not present in the blood stream in large numbers until just before death, a blood specimen is of no value early in the disease.

ASCARIASIS

1. *Etiologic agent. Ascaris lumbricoides,* the large intestinal roundworm of man.

2. *Source of infection.* Infective ova from human feces, particularly those of children, deposited in and about houses where facilities for sanitary disposal of human excreta are lacking or not used.

3. *Mode of transmission.* By direct or indirect transmission of the embryonated eggs from soil or other contaminated material to the mouth. Salads may be contaminated. The embryonated eggs hatch in the intestinal canal, and the larvae penetrate the wall, and reach the lungs by way of the lymphatic and the circulatory systems. Most of those which reach the lungs pass into the air passages, ascend the bronchi, are swallowed and eventually reach the small intestine where they grow to maturity. Contaminated soil may be carried long distances on the feet or footwear into houses and conveyances.

4. *Prevalence.* High incidence of infection is found where low standards of hygiene, lack of sanitary essentials, poverty, and ignorance create the conditions conducive to intensive contamination of soil in the immediate vicinity of houses. Children of the runabout and early school age are likely to be more frequently and more heavily infected than are older children and adults. Particularly prevalent in the United States among the people of the Appalachian plateau.

5. *Current laboratory services.* Microscopic examination of feces for ova of *Ascaris lumbricoides* or identification of adult worm when passed.

6. *Collection of specimens.* With a tongue depressor, spoon, or other instrument collect specimen of feces, preferably from different parts of the stool, and place in bottles contained in "PD" outfit.

7. *Limitations of laboratory tests.* A series of stool specimens should be submitted for examination for intestinal parasites before placing too great reliance upon negative findings. The finding of ova is dependent upon the presence of female worms in the intestine.

BARTONELLOSIS (OROYA FEVER, VERRUGA PERUANA)

1. *Etiologic agent. Bartonella bacilliformis.*

2. *Source of infection.* The blood of an infected individual.

3. *Mode of transmission.* Sand flies of the genus *Phlebotomus.*

4. *Prevalence.* Known only at certain altitudes in mountain valleys on the Pacific slope in Peru, Ecuador and Columbia.

5. *Current laboratory services.* Examination of blood smears for organisms morphologically typical of *Bartonella bacilliformis.*

6. *Collection of specimens.* Make thin blood films as for differential white cell count using slides in "MA" outfit. Allow to dry without application of heat. Mark history blank plainly "For *Bartonella*".

7. *Limitations of laboratory tests.* The findings of morphologically typical organisms in blood films is considered confirmatory of clinical evidence of this infection. Specimens taken at the height of the fever are the most satisfactory.

BOUTONNEUSE FEVER (MEDITERRANEAN FEVER)

1. *Etiologic agent. Rickettsia conori.*

2. *Source of infection.* Infected ticks, dogs and perhaps rodents.

3. *Mode of transmission.* Bite of an infected tick, *Rhipicephalus sanguineus,* which infests dog kennels and households where dogs are quartered.

4. *Prevalence.* The disease has been reported from the entire Mediterranean coast line area, Rumania and Portugal. Similar diseases which may be identical with boutonneuse fever have been reported from Kenya and South Africa (tick typhus). The disease has not been recognized in the United States.

5. *Current laboratory services.* Serological tests.

6. *Collection of specimens.* Use "MI" outfit for collection of whole blood. Collect two specimens, one early in the disease and the other during the third or fourth week of illness.

7. *Limitations of laboratory tests.* The Weil-Felix agglutination titer is reported to be lower than in other rickettsial infections. The rise in titer between the first and second specimens has some value but does not indicate which rickettsial infection is present.

BRUCELLOSIS (UNDULANT FEVER)

1. *Etiologic agent. Brucella melitensis; Brucella abortus; Brucella suis.*

2. *Source of infection.* The tissues, blood, milk and dairy products, and urine of infected animals, especially goats, cattle and swine. Laboratory infections occur frequently.

3. *Mode of transmission.* By ingestion of milk or dairy products from infected animals and by direct contact with infected animals or animal products.

4. *Prevalence.* Occurs more often in males than in females, and particularly in persons whose occupation brings them into direct contact with milk, cows, hogs or goats, and in persons using unpasteurized milk of cows or goats. Found throughout the United States and Canada, affecting persons of all races usually as sporadic cases but occasionally in small epidemics. In the Midwestern states *Brucella suis* is of great importance in human infections but *Brucella melitensis* infections are by no means uncommon. Occurs most often in the months of May to October. Latent or missed cases far outnumber reported cases.

5. *Current laboratory services.*

   A. Agglutination tests on blood specimens.

   B. Blood cultures for *Brucella;* cultures routinely made on clots from blood specimens submitted for agglutination tests.

   C. Special studies on suspected sources of infection available by special arrangement.

6. *Collection of specimens.*

   A. **For agglutination tests and clot cultures:** Use "TY" outfit. After the 10th day of illness, collect aseptically 5-10 ml. of venous blood and place in sterile bottle in outfit. In acute cases, follow up with a specimen collected 3-4 weeks after onset.

   B. **Blood cultures:** Use "BC" outfit. During febrile episode, prepare the skin at the bend of the elbow with iodine and alcohol. Place a tourniquet on the arm. Have the patient extend the arm fully and open and close the fist a few times to distend the veins. With the sterile syringe and needle, remove not less than 5 ml. Have the patient open his fist, then remove the tourniquet and withdraw the needle. Preferably replace used needle with another unused sterile one. To transfer blood to the culture outfit pass the needle of the syringe through the rubber diaphragm in the top of the screw cap (foil covering having been removed previously), and force the blood directly into the bottle. The enriched medium in the container will prevent clotting of blood and support growth of organisms that may be present. It is important that not less than 5 ml. or more than 10 ml. of blood be added. The outfit should be returned to the laboratory as soon as possible.

   C. **Milk and similar materials:** Contact Bureau of Laboratories for special instructions before sending any specimens or samples.

7. *Limitations of laboratory tests.*

   A. **Agglutination tests:** Strong agglutination is highly suggestive and confirmatory of suspected brucellosis. Weak reactions may have significance in many cases. Some infected

persons do not show detectable agglutinins; hence, negative findings are not conclusive. In acute cases, a rise in titer in serial specimens is of significance. Some cross-agglutination in serum from tularemia cases or from individuals who have received cholera vaccine.

B. **Blood cultures and clot cultures:** Since *Brucella* are present in the blood stream in detectable numbers only during unpredictable "showers" and since this is most likely to occur during febrile episodes, negative results on a single specimen are not conclusive. A series of specimens is recommended. Positive findings are diagnostic.

C. **Special tests on milk, etc.:** Demonstration of agglutinins in milk serum or isolation of the organism from milk or similar products provides evidence of epidemiological value only.

CESTODE INFECTIONS, MISCELLANEOUS*

(TAPEWORMS)

1. *Etiologic agent.* Among others are *Taenia saginata* (beef tapeworm), *Taenia solium* (pork tapeworm), *Hymenolepis nana* (dwarf tapeworm), *Dipylidium caninum, Diphyllobothrium latum* (broad Russian tapeworm).

2. *Source of infection.* Infected humans or animals.

3. *Mode of transmission.* An intermediate host is required except for *Hymenolepis* where transmission may be direct. Infection occurs by ingestion of the eggs (direct) or of cysticerci in incompletely cooked flesh of intermediate hosts.

4. *Prevalence.* Widespread in the United States.

5. *Current laboratory services.* Microscopic examinations of feces for ova, and of proglottids when found.

6. *Collection of specimens.* Use "PD" outfit. Place portions of stool (scolex or proglottids when present) in bottle provided.

7. *Limitations of laboratory tests.* Identification of species from the ova is possible except for *Taenia solium* and *Taenia saginata* which cannot be distinguished from each other except by study of scolex or proglottids.

*See also "Echinococcosis (Hydatid Disease)".

CHANCROID (SOFT CHANCRE)

1. *Etiologic agent.* Ducrey bacillus, *Hemophilus ducreyi.*

2. *Source of infection.* Discharges from lesions.

3. *Mode of transmission.* Chancroid is predominantly venereal in origin but has occurred rarely on the hands of doctors and nurses through professional contact with infected persons. It may similarly occur in children through accidental inoculation. Acquisition through indirect contact with articles soiled with moist discharges from the lesions of infected persons is rare.

4. *Prevalence.* Geographically widespread and particularly common among populations sexually promiscuous and living at a low economic and social level.

5. *Current laboratory services.*

   A. Exclusion procedures:

      a. Dark field examination for *Treponema pallidum* of syphilis.

      b. Blood test for syphilis.

      c. Blood test for lymphogranuloma venereum.

   B. Microscopic examination of exudate from lesions, pus from buboes, etc.

   C. Cultural examination of exudate from lesions, pus from buboes, etc.

6. *Collection of specimens.*

   A. See directions under "SYPHILIS" and "LYMPHOGRANULOMA VENEREUM".

   B. Use "GC" or "VI" outfit. With sterile swab furnished collect material from lesion and smear on microscope slides. Mark history blank plainly "For chancroid".

   C. Use "HS" outfit. With sterile swab collect deep exudate from lesion or aspirated pus from unruptured bubo. Then plunge swab into jelly in tube and replace in outfit for mailing. Mark history blank plainly "for chancroid".

7. *Limitations of laboratory tests.*

   A. Because direct laboratory aids (B and C) to the diagnosis of chancroid are not always successful, the exclusion of syphilis and of lymphogranuloma venereum is an important diagnostic procedure.

   B. and C. These measures are successful only in 50-75 per cent of cases. The finding of typical organisms is considered confirmatory of clinical evidence of this infection. Failure to demonstrate the organism is not a reliable criterion for exclusion of this infection.

CHICKENPOX (VARICELLA)

1. *Etiologic agent.* The virus of chickenpox (*Briareus varicellae*).

2. *Source of infection.* The infectious agent is present in the lesions of the skin and presumably of the respiratory tract, which may render the disease communicable before the exanthem is in evidence.

3. *Mode of transmission.* Directly from person to person; indirectly through articles freshly soiled by discharges from the skin and mucous membranes of infected persons.

4. *Prevalence.* Universal. Probably 70 per cent of persons have had the disease by the time they are 15 years of age. Not uncommon in early infancy. Winter and spring are seasons of greatest prevalence in North America.

5. *Current laboratory services.* No practicable laboratory aids to diagnosis.

CHOLERA (ASIATIC CHOLERA)

1. *Etiologic agent.* Cholera vibrio, *Vibrio comma,* commonly subtypes Inaba and Ogawa.

2. *Source of infection.* Feces and vomitus of patients, feces of convalescents, persons with inapparent infections, and carriers. The carrier state is temporary.

3. *Mode of transmission.* By direct or indirect fecal contamination of water or of foods by soiled hands, utensils or flies.

4. *Prevalence.* The endemic center of this disease is in India and certain areas of southeastern Asia. From these centers it spreads along lines of human communication, from time to time reaching more remote countries, and causing widespread epidemics. Not endemic in the Western Hemisphere.

5. *Current laboratory services.* Cultural and microscopic examination of feces for *Vibrio comma.*

6. *Collection of specimens.* There is a federal prohibition against admission of cholera specimens to the mails. Specimens must be sent by messenger. Collect one or two ml. of the feces which are usually liquid ("rice-water stools") and place in the sterile bottle in the "FE" outfit.
Mark history blank plainly "For cholera".

7. *Limitations of laboratory tests.* A few organisms that closely resemble *Vibrio comma* are differentiated from it with difficulty.

COCCIDIOIDOMYCOSIS* (COCCIDIOIDAL GRANULOMA, "VALLEY FEVER")

1. *Etiologic agent. Coccidioides immitis.*

2. *Source of infection.* Dust, soil and vegetation contaminated with the spores of the fungus.

3. *Mode of transmission.* Inhalation of spores in dust and dry vegetation and, in laboratories, inhalation of spores from cultures. Infection through skin wounds is a possible but infrequent route.

4. *Prevalence.* Known endemic areas are southwestern United States, northern Mexico and the Chaco region of Argentina and Uruguay. Incidence highest in hot dry weather. Travel through an endemic area may result in infection but that is rare.

5. *Current laboratory services.*

   A. Microscopic and cultural examination of material from lesions.

   B. Microscopic and cultural examination of sputum in the pulmonary type of infection.

   C. Serologic test on blood serum.

6. *Collection of specimens.*

   A. Collect pus in "FI" outfit for cultural and microscopic examination.

   B. Collect sputum that has been coughed up from the lungs. Place sputum in sterile bottle, preferably in "MI" outfit. (If "TC" or "PN" outfit is used, history blank must be plainly marked "For coccidioidomycosis".)

   C. Collect aseptically 10 ml. of venous blood and place in sterile bottle in "MI" outfit. Mark history blank plainly "For serologic test for coccidioidomycosis".

7. *Limitations of laboratory tests.* Suggestive microscopic findings should be confirmed by cultural identification of the organism, *Coccidioides immitis,* and by animal inoculation. Serologic tests for coccidioidomycosis are still experimental; the findings must be weighed carefully in the light of clinical evidence and history. (Intradermal tests with "coccidioidin", an antigen prepared from the fungus, may be helpful.)

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*Also called "Desert Rheumatism", "San Joaquin Fever", "Posada-Wernicke Disease", "Coccidioidal Erythema Nodosum".

COMMON COLD

1. *Etiologic agent.* One or more viruses (*Tarpeia premens).*
2. *Source of infection.* Discharges from nose and mouth of infected persons.
3. *Mode of transmission.* Usually directly by coughing, sneezing and explosive manner of speech by which droplets pass in the air from the infected person to susceptible persons, especially within short range; and indirectly by handkerchiefs, eating utensils, or other articles freshly soiled by discharges of the infected person.
4. *Prevalence.* Most persons, except those living in small isolated communities, have one or more colds each year. The incidence does not vary materially according to age, sex, race or occupation, but incidence is higher in children under five years of age and becomes less after 20 years.
5. *Current laboratory services.* No specific diagnostic laboratory aid available.

CONJUNCTIVITIS, ACUTE INFECTIOUS*

(OF THE NEW BORN, NOT INCLUDING TRACHOMA)

1. *Etiologic agent.* The gonococcus (*Neisseria gonorrheae)* is the most important but not necessarily the most frequent infecting micro-organism. Others include meningococcus (*Neisseria meningitidis),* hemophilic bacilli and a virus (inclusion blenorrhea).
2. *Source of infection.* Discharges from conjunctivae, or adnexa, or genital mucous membranes of infected persons.
3. *Mode of transmission.* Contact with an infected person or with articles freshly soiled with discharges of such person.
4. *Prevalence.* Occurrence varies widely according to the observance or neglect of prophylactic use of a solution of silver nitrate or equivalent preparation in the eyes of the newborn by the attendant at the delivery. An infrequent complication in the present-day care of the new born.
5. *Current laboratory services.* Microscopic and cultural examinations of pus or exudate for identification of organism, especially for presence or absence of gonococcal infection.
6. *Collection of specimens.* Submit smear of pus or exudate on slides in "GC" outfit. Material for general culture may be submitted on sterile swab in "HS" outfit; mark history blank "For identification of organism".

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*Includes gonorrheal ophthalmia, ophthalmia neonatorum, and babies' sore eyes in the first 21 days of life.

7. *Limitations of laboratory tests.* The finding of gram-negative intracellular diplococci is strong presumptive evidence of gonococcal infection. Isolation of common pyogenic bacteria may result from skin contamination. Certain forms of this disease are apparently of viral etiology (inclusion blenorrhea) for which no specific laboratory test is available.

DENGUE

1. *Etiologic agent.* The virus of dengue fever; at least two immunologically distinct strains have been identified.
2. *Source of infection.* The blood of infected persons one day before and up to 5 days after onset.
3. *Mode of transmission.* By the bite of mosquitoes, *Aedes aegypti, Aedes albopictus* or *Aedes scutellaris* infected by biting a patient. The mosquito becomes infectious after an interval of 8-11 days.
4. *Prevalence.* May occur wherever the mosquito vectors exist, mainly in the tropics and subtropics.
5. *Current laboratory services.* No diagnostic laboratory aid available at the Bureau of Laboratories.
6. *Collection of specimens.* Contact Bureau of Laboratories before submitting any specimen. Providing arrangements can be made for submission to a laboratory where immunological studies are being carried on for this disease, fresh specimens of blood should be drawn aseptically without preservative or anticoagulant, (1) during the acute stage and (2) during the convalescent stage.
7. *Limitations of laboratory tests.* At present the lack of facilities for specific diagnostic aids constitutes the main limitation. A rise in the neutralizing power of the patient's serum against the virus during convalescence is strong presumptive evidence of the disease.

DIARRHEA OF THE NEW BORN, EPIDEMIC

1. *Etiologic agent.* Unknown.
2. *Source of infection.* Unknown.
3. *Mode of transmission.* Unknown, presumably direct or indirect person to person infection.
4. *Prevalence.* The disease is met with frequently in North America and Europe and is probably more widely spread. No definite seasonal incidence has been demonstrated.
5. *Current laboratory services.* No specific diagnostic laboratory aids available at Bureau of Laboratories but cultural examination of feces specimens for the exclusion of known pathogens is recommended.

6. *Collection of specimens.* For purposes suggested above under *"Current laboratory services"* submit specimens of feces in "FE" outfit. Using the sterile swab in the outfit, transfer a portion of the stool about the size of a sphere ½" in diameter to the specimen jar and emulsify it in the glycerol solution. If stools are liquid, transfer to the specimen jar with a clean spoon or other implement a quantity approximately 2 ml. in volume. Do not use spoon or other implement for more than one sampling unless carefully sterilized in the interim.

7. *Limitations of laboratory tests.* For the present, no specific laboratory test is available; tests must be confined to those for the exclusion of known bacterial pathogens.

DIPHTHERIA

1. *Etiologic agent. Corynebacterium diphtheriae* (the Klebs-Loeffler bacillus).

2. *Source of infection.* Discharges and secretions from surfaces of nose, throat and nasopharynx of infected persons and carriers of the bacillus and from skin lesions.

3. *Mode of transmission.* Association with a case or a carrier or with articles soiled with the discharges of such persons. Milk has served as a vehicle.

4. *Prevalence.* Endemic and epidemic. The disease is more common in temperate zones than elsewhere, especially in fall and winter months. Approximately one-quarter of the cases and one-half of the deaths occur in children under five years of age. Age distribution of cases and deaths depend much on the state of immunization of the child population.

5. *Current laboratory services.*

   A. Cultural examination for *Corynebacterium diphtheriae;* typing of organisms isolated.

   B. Determination of virulence (toxigenicity) of *Corynebacterium diphtheriae* by animal inoculation tests.

6. *Collection of specimens.*

   A. For cultures: Use "KL" outfit. Rub the sterile swab marked "Throat" gently but firmly against affected area; if "false membrane" is present, collect swabbings from the margin or just beneath the membrane. When no such membrane is present, rub the swab over the mucosa of the lower pharynx and tonsils before inoculating throat culture tube. Bacterial contamination may result if small food particles are picked up by the swab. Under no conditions should any attempt be made to collect material shortly after application of antiseptics or germicides to the throat.

Then rotate swab gently over the coagulated blood serum medium in the vial marked "Throat" but do not break the surface of the medium. Do not discard the swab but replace it in the envelope and return it in the outfit with the culture. Then rub the sterile swab marked "Nose" gently over the mucous membrane of the nasal cavities. Cultures from the nostrils are more successful if the nostrils are first cleansed with physiological salt solution. Inoculate culture tube marked "Nose" and return swab as directed above.

When suspected diphtheritic skin lesions are present, collect swabbings directly from lesions, inoculate throat or nose tube as above and mark history blank plainly "skin culture".

B. Virulence tests are performed on organisms isolated from specimens to confirm cultural findings.

7. *Limitations of laboratory tests.*

A. A positive report on a culture means that organisms morphologically typical of the diphtheria bacillus were observed. This is considered confirmatory of clinical evidence of the disease; the organisms may also be found in carriers who are not ill. A single negative culture should not be considered conclusive if there are clinical symptoms. In all such cases another culture from the throat and also one from the nose should be sent. Negative reports on at least two successive cultures taken at least 24 hours apart from nose and throat are required for releasing contacts and cases of diphtheria.
(Typing of diphtheria organisms as *gravis, mitis* or *intermedius* has no diagnostic significance except to establish that the organism is a true diphtheria organism and not a similar organism of no significance. It is considered of some epidemiological value when diphtheria is prevalent.)

B. A report that a culture is virulent means that the Bureau of Laboratories has determined that the culture produced gross pathology in the guinea pig typical of that produced by diphtheria toxin and that an animal protected by antitoxin was not similarly affected.

DYSENTERY, BACILLARY (SHIGELLOSIS)

1. *Etiologic agent.* Various species of the genus *Shigella.*

2. *Source of infection.* The feces of infected persons and carriers. Healthy carriers are common.

3. *Mode of transmission.* By eating contaminated foods, or drinking contaminated water or milk and by hand-to-mouth transfer of contaminated material; by flies; from objects soiled with discharges of a patient or of a carrier.

4. *Prevalence.* Endemic, epidemic and sporadic, but shares with other enteric infections the characteristics of a striking and progressive reduction wherever water supplies are rendered safe, sewage is disposed of in a sanitary manner, milk is pasteurized and infant hygiene is of a good order. Most common in the summer months. Institutional outbreaks are frequent.

5. *Current laboratory services.* Cultural examinations of feces for the various types of dysentery bacilli. Facilities are also available for serological typing of cultures isolated at other laboratories which lack typing facilities. Agglutination tests are not recommended (See below).

6. *Collection of specimens.* Use "FE" outfit. Using the sterile swab in the outfit, transfer a portion of the stool about the size of a sphere ½" in diameter to the specimen jar and emulsify it in the glycerol solution. If stools are liquid, transfer to the specimen jar with a clean spoon or other implement a quantity approximately 2 ml. in volume. Do not use spoon or other implement for more than one sampling unless carefully sterilized in the interim. Care should be taken that the stool is passed into a vessel that has been thoroughly disinfected or sterilized.

7. *Limitations of laboratory tests.* Final identification of dysentery bacilli depends upon adequate serological tests. Unless such tests are performed, dysentery-like organisms may be mistaken for those which cause bacillary dysentery. The type reported is of importance in establishing relationship to other cases or to carriers of dysentery organisms. The examination of blood specimens for agglutinins has been discontinued by the Bureau of Laboratories because many persons, particularly adults, may show confusingly high titres without evidence of infection. Moreover, the development of agglutinins in infected persons is slow and inconsistent.

ECHINOCOCCOSIS (HYDATID DISEASE)

1. *Etiologic agent. Echinococcus granulosus* (*Taenia echinococcus*), an intestinal parasite of dogs.

2. *Source of infection.* Infected dogs and contaminated animal products, hides, wool, etc., of herbivora, and contaminated environment.

3. *Mode of transmission.* Hydatid disease is transmitted from dog to man by ova present on tongue and hair, through handling and contact. Domestic herbivorous animals are infected by drinking water or eating food contaminated with dog feces. Ova of *Echinococcus granulosus* are very resistant to outside temperatures (freezing and summer heat) and to drying, and may be carried by wind, dust or flies .

4. *Prevalence.* Comparatively infrequent in the United States. Principal foci in the Americas are in Argentina, Uruguay and Chile. Known in many parts of the world.

5. *Current laboratory services.*

   A. Serological tests on blood specimens available through special arrangement with cooperating laboratories.

   B. Examinations of cyst fluid for scolices of *Echinococcus granulosus.* (Exploratory aspiration not recommended.)

6. *Collection of specimens.*

   A. Use "MI" outfit. Collect aseptically 5-10 ml. of blood and place in sterile bottle in outfit. Mark history blank plainly "For echinococcosis".

   B. When cysts are encountered at autopsy or during operation, place fluid in bottle in "MI" outfit; mark history blank plainly "For echinococcosis".

7. *Limitations of laboratory tests.*

   A. A reaction to the serological test is confirmatory of clinical evidence. Negative results are not entirely conclusive.

   B. The examination of cyst fluid for scolices is of limited value since exploratory aspirations of the fluid is a dangerous procedure and securing of material is usually limited to autopsy specimens or specimens taken at operation. (Intradermal tests are also recommended.)

ENCEPHALITIS, INFECTIOUS

(SEE ALSO LYMPHOCYTIC CHORIOMENINGITIS)

A. Encephalitis Lethargica (Vienna or von Economo type).

1. *Etiologic agent.* Unknown.

2. *Source of infection.* Unknown.

3. *Mode of transmission.* Unknown.

4. *Prevalence.* Worldwide from about 1918 through 1926 in endemic or epidemic form, seen only occasionally at present. Possibly these cases seen now are of different etiology. Was prevalent particularly in winter months and highest incidence was in young persons.

5. *Current laboratory services.* None except for exclusion of other known encephalitides (See B and C below).

B. Arthropod-borne Virus Encephalitides

1. *Etiologic agents.* Each form of the disease is caused by a specific virus — the equine types (Eastern, Western and Venezuelan) by *Erro equinus,* the Japanese B type by *Erro japonicus,* the St. Louis type by *Erro scelestus,* Russian (Siberian), spring-summer

type by *Erro silvestris,* louping ill by *Erro scoticus,* West Nile type (known cases were symptomless) by *Erro nili.* Some show a slight immunological cross-relationship.

2. *Source of infection.* Birds, wild and domestic, serve as the principal source of mosquito infection in the United States. In certain areas of North America mites of chickens and wild birds are found infected, and through transovarian infection are suspected of serving as permanent reservoirs. The equine species, though serving as host, is not apparently important as a source of mosquito infection. Man probably not a source of infection for types found in United States.

3. *Mode of transmission.* Mosquitoes for all but Russian spring-summer type and louping ill which are tick-borne and for West-Nile type about which little is known. In western and central United States and in Canada, *Culex tarsalis* is believed to be the principal vector. Genus and species in the East and elsewhere are unknown.

4. *Prevalence.* In the United States a disease of summer and early fall, limited to areas and years of high sustained temperature and large numbers of mosquitoes. Endemic in the United States in many hot, irrigated western valley areas, but irregularly epidemic in dry farming areas of the midwest, southwest and east. Highest rates in rural and suburban districts.

5. *Current laboratory services.* Complement-fixation tests for equine, Japanese B and St. Louis types; also for other viral encephalitides (See C below).

6. *Collection of specimens.* Use "MI" outfit. Collect 20 ml. of blood and allow to clot. Mark history blank "For encephalitides". Always submit two specimens, one during acute illness and one 3-4 weeks later.

7. *Limitations of laboratory tests.* It is essential that a rise in titer be demonstrated in comparing results of the convalescent stage test with that made during the acute phase. Negative results do not rule out the possibility of an infectious encephalitis due to some virus not recognized.

C. Other Virus Encephalitides

1. *Etiologic agent.* Numerous viruses such as those of mumps (*Rabula inflans),* lymphogranuloma venereum (*Miyagawanella lymphogranulomatis),* herpes simplex (*Scelus recurrens),* etc.

2. *Source of infection.* The same as the sources for the more common syndromes caused by these viruses.

3. *Mode of transmission.* The same as for transmission of these viruses when they cause the more usual manifestations of disease.

4. *Prevalence.* Central nervous system involvements in infections with these viruses are infrequent except for mumps meningoencephalitis which is encountered with higher frequency usually during epidemics.

5. *Current laboratory services.* Complement-fixation tests for mumps and lymphogranuloma venereum.

6. *Collection of specimens.* Use "MI" outfit. Collect 20 ml. of blood and allow to clot. Mark history blank "For encephalitis". Always submit two specimens, one during the acute stage and one 3-4 weeks later.

7. *Limitations of laboratory tests.* It is essential that a rise in titer be demonstrated in comparing results of the convalescent stage test with that made during the acute phase. Negative results do not rule out the possibility of an infectious encephalitis due to some virus not recognized.

ENTEROBIASIS (OXYURIASIS, PINWORM OR THREADWORM INFECTION)

1. *Etiologic agent. Enterobius vermicularis (Oxyuris vermicularis),* an intestinal roundworm parasitizing only mankind. The ovum is infective within a few hours after leaving the gastrointestinal tract.

2. *Source of infection.* Infected persons, particularly children; clothing and bedding soiled with ova.

3. *Mode of transmission.* Infective ova may be transmitted directly by hand from the anal region to the mouth of the same host and indirectly to the same host or to new hosts through contamination of food and other objects. Dust-borne infection is common in contaminated households. After ingestion, the ova hatch in the stomach and small intestine, cecum, and upper portion of the colon. The gravid worms migrate to the rectum to discharge their ova on the perianal skin. In female patients they may migrate up the genital system and enter the peritoneal cavity.

4. *Prevalence.* Cosmopolitan. It is estimated that about 20 per cent of the general population of the United States are infected. The incidence is highest in children of school age, next highest in those of preschool age, and lowest in adults except among mothers of infected children among whom the rate is commonly high. Infection is characteristically familial, and crowding is an important factor. Incidence of infection is often high in institutional environments.

5. *Current laboratory services.* Examination of anal swabbings for ova of *Enterobius (Oxyuris) vermicularis;* identification of adult worms passed in feces.

6. *Collection of specimens.* Use "PW" outfit. Collect specimens in the morning only and immediately after patient has awakened.

To collect eggs deposited by worms during patient's sleep, use swab furnished in the tube as directed by gently stroking the anus and adjacent area; return swab to tube and submit to the laboratory. For submission of adult worms use "PD" outfit.

7. *Limitations of laboratory tests.* Examination of anal swabbings is more reliable than examination of stool specimens because the eggs are actually deposited outside the anus by the gravid female of *Enterobius.*

FAVUS*

1. *Etiologic agent. Trichophyton schoenleini (Achorion schoenleini).*
2. *Source of infection.* Lesions of skin, particularly of scalp, rarely of nails.
3. *Mode of transmission.* Direct contact with patient, and indirectly through toilet articles.
4. *Prevalence.* Rare in children in North America, and when occurring can usually be traced to immigrants from southern and eastern Europe. Tends to spread within families.
5. *Current laboratory services.* Microscopic and cultural examinations of infected hairs or skin scrapings for *Trichophyton schoenleini (Achorion schoenleini).*
6. *Collection of specimens.* Use "FI" outfit; select fluorescent hairs using a Wood's lamp, remove from scalp with sterile forceps and place a few, well separated, on surface of agar medium. Select a few more and place between the glass slides and wrap tightly. When skin scrapings are submitted, first cleanse area with 70% alcohol, then remove scrapings with sterile instrument and proceed as directed for hairs.

FILARIASIS

1. *Etiologic agent.* A nematode worm, *Wuchereria bancrofti* or *malayi.* Several other species of filariids are known to infect man; serious clinical disease is most commonly caused by *Wuchereria bancrofti.*
2. *Source of infection.* The blood of an infected person.
3. *Mode of transmission.* In North America has been transmitted by the mosquito *Culex quinquefasciatus.* Other mosquitoes including representatives of the genera *Culex, Aedes, Anopheles* and *Mansonia* have been incriminated in other parts of the world. After the mosquito takes a blood meal from a person with circu-

*Also known as Tinea Favosa, Favus Honeycomb Ringworm. See also "Ringworm of the Scalp."

lating filaria embryos, the embryos develop in the mosquito into infective larvae in 14 to 21 days. Transmission is during the feeding of the mosquito.

4. *Prevalence.* Not transmitted in the continental United States; previously reported cases were limited to Charleston, S.C. This focus of infection no longer exists. Common in many tropical and sub-tropical regions. The infection is not naturally acquired in continental United States.

5. *Current laboratory services.*

   A. Microscopic examination of blood for microfilaria,

   B. Microscopic examination of fluid from lymph node puncture, hydrocele or similar material for microfilaria.

   C. Serological tests on blood specimens are not made in Bureau of Laboratories but available by arrangement with research laboratories. (Examination of biopsy material by a skilled pathologist is also recommended by some authorities.)

6. *Collection of specimens.*

   A. Collect thick blood films as for malaria examination. Use "MA" outfit. Follow directions given under "MALARIA". Late evening specimens may be necessary and concentration methods may be advisable with filaria exhibiting periodicity. Before submitting fluid blood for concentration, contact Bureau of Laboratories and a special outfit (containing 9 ml. of 2% formalin) will be provided. In either case plainly mark history blank "For filaria".

   B. Collect fluid in sterile bottle in "MI" outfit; mark history blank plainly "For filaria".

   C. Collect 5-10 ml. blood in sterile bottle in "MI" outfit; mark history blank plainly "For serological test for filariasis".

7. *Limitations of laboratory tests.*

   A and B. Identification of microfilaria is indication of infection. Since certain filaria exhibit periodicity both day and night blood should be examined. In any case, negative results are not conclusive.

   C. Serological tests for filariasis with antigen from filaria of the dog (*Dirofilaria immitis)* are said to be specific.

FOOD POISONING: BACTERIAL INTOXICATIONS

A. Staphylococcus

1. *Etiologic agent.* Toxin (enterotoxin) of certain strains of staphylococci (*Micrococcus pyogenes,* usually variety *aureus;* more commonly known as *Staphylococcus aureus).* Toxin is stable at boiling temperature; staphylococci multiply in food, producing a toxin which is the cause of poisoning.

2. *Source of infection.* (Not an infection but a poisoning). Source of contamination not known in most cases, believed to be of human origin.

3. *Mode of transmission.* Most common vehicle is custard-filled pastry; processed meats, especially ham, responsible for some outbreaks; outbreaks reported due to milk from cows with specifically infected udders.

4. *Prevalence.* Widespread; probably the principal cause of "food poisoning".

5. *Current laboratory services.* Examinations of foods for enterotoxigenic staphylococci and for enterotoxin.

6. *Collection of specimens.* Rush suspected foods to laboratory in suitable container; accompany by description of symptoms and probable incubation period.

7. *Limitations of laboratory tests.* The enterotoxin is preformed by bacterial growth in the foods. Hence, stool cultures are not recommended except when *Salmonella* infections must be excluded. In general food poisoning occurring within eight hours (usually 2-4 hours) after ingestion of the suspected food support the probability of illness due to staphylococcus enterotoxin as opposed to *Salmonella* infections although relatively short incubation periods (6-12 hours) for the latter have been known. See "SALMONELLOSIS". A negative test may result when unaffected portions of the food are all that remain for sampling.

B. Botulinus (Botulism)

1. *Etiologic agent.* The toxins produced by the botulinus bacillus (*Clostridium botulinum, C. parabotulinum)* in food improperly processed. Toxin is produced only under anaerobic conditions, especially in non-acid foods. Toxin easily destroyed by boiling but spores require higher temperatures for destruction.

2. *Source of infection.* (Not an infection but a poisoning). Spores of botulinus bacillus occur in soil and have been found in the intestinal tract of certain animals. Toxin is formed by anaerobic growth in food.

3. *Mode of transmission.* Food containing the botulinus toxin, usually eaten from jars or cans inadequately processed during canning. Most poisonings are due to home canned vegetables.

4. *Prevalence.* Sporadic cases and groups of cases occur in all countries and always in relation to some perishable food product which has been so kept or preserved as to permit the development, under partially anaerobic conditions, of *Clostridium botulinum,* or of *Clostridium parabotulinum* to the extent of forming the toxin that causes the symptoms. In the United States the disease has in recent years followed most commonly the use, without further or adequate cooking, of home-canned vegetables and meat products.

5. *Current laboratory services.* Examination for toxin of *Clostridium botulinum.*

6. *Collection of specimens.* Rush suspected foods to laboratory in suitable container; accompany by description of symptoms and probable incubation period; state if botulism is suspected.

7. *Limitations of laboratory tests.* Successful demonstration of the toxin is dependent upon selection of samples of food most likely to have supported the growth of the organism which grows only in the absence of air and does not usually grow in acid fruits and vegetables. Home-canned products are most likely sources.

GAS GANGRENE (ANAEROBIC WOUND INFECTIONS)

1. *Etiologic agent.* Various anaerobic spore-forming bacilli including *Clostridium perfringens* and many other species.

2. *Source of infection.* The organisms are widely distributed in nature but their main habitat is the soil. Some are common intestinal inhabitants of man and other animals.

3. *Mode of transmission.* By introduction of organisms into deep wounds at time of injury or by subsequent contamination.

4. *Prevalence.* Although the organisms are widespread in nature they require dead tissue and anaerobic conditions for growth and toxin production; hence, the incidence is kept low by modern asepsis and surgical technic.

5. *Current laboratory services.* Bacteriological examinations of pus or curettings from wound.

6. *Collection of specimens.* Use "MI" outfit. Place material in sterile bottle in outfit. Mark history blank plainly "For anaerobic wound infection".

7. *Limitations of laboratory tests.* Only those spore-forming organisms (members of the genus *Clostridium)* which are obligatory anaerobes are significant.

GLANDERS

1. *Etiologic agent.* The glanders bacillus, *Malleomyces mallei* (*Bacillus mallei*).

2. *Source of infection.* Discharges from open lesions of mucous membranes or of the skin of infected horse or man (e.g., pus and mucous from the nose or throat, or bowel discharges).

3. *Mode of transmission.* Contact with infected horse or man or with articles freshly soiled by discharges therefrom.

4. *Prevalence.* Rare and sporadic and almost exclusively in men occupied about horses. In widespread and local epidemics as an epizootic in horses. Infrequent in the United States.

5. *Current laboratory services.* Cultural examinations of pus or exudates from lesions for *Malleomyces mallei.* Arrangements for serological tests of blood, particularly from animal sources, can be made when needed.

6. *Collection of specimens.* Contact Bureau of Laboratories before collecting specimens. Special instructions will be given.

7. *Limitations of laboratory tests.* Cultural identification of the organism is confirmatory of glanders suspected clinically. Serological tests are of value after duration of several weeks only, occasionally during the second week.

GONORRHEA (INCLUDING SEQUELAE OF GONOCOCCAL ETIOLOGY)

1. *Etiologic agent.* Gonococcus, *Neisseria gonorrheae.*

2. *Source of infection.* Discharges from lesions of inflamed mucous membranes and lymph nodes of infected persons.

3. *Mode of transmission.* By direct personal contact with infected persons, and rarely by indirect contact with articles freshly soiled with the discharges of such persons. In adults by sexual intercourse; in children by personal and indirect contact with discharges; in the new born by ophthalmic infection at birth.

4. *Prevalence.* Widespread. Occurs among both sexes and at all ages, but is most prevalent among persons in the age groups of greatest sexual activity.

5. *Current laboratory services.*

   A. Microscopic examination of smears.

   B. Complement-fixation tests as aids to the diagnosis of obscure conditions of possible gonococcal etiology (such as gonococcal arthritis) are available in limited numbers by arrangements with cooperating laboratories.

6. *Collection of specimens.*

A. Smears:

Use "GC" outfit. Make at least two smears. Mark or number each to indicate whether from urethra, cervix or other source. In the male take both smears from the urethral discharge (unless prostatic smears are preferred). In the female, take one from the urethral discharge, if there is any, and the other from the cervix uteri. Do not collect the discharge from between the labia or from the vagina as it is unsatisfactory for microscopic examination. In both sexes milk the urethra before making the smears. Make a thin smear from the discharge on one of the slides with one of the swabs, and a second smear on the other slide with the other swab. Let each smear dry at room temperature before putting the slides together. (Failure to observe this precaution makes a reliable examination impossible.) Diagnosis of specific vaginitis in children should always be confirmed by culture.

B. Complement fixation test:
Submit about 5 ml. of blood in bottle in "MI" outfit. Mark history blank plainly "For GC complement fixation".

7. *Limitations of laboratory tests.*

A. Smears:
Results reported as positive are based on the finding of gram-negative intracellular diplococci morphologically identical with *Neisseria gonorrheae.* A positive result means that the microscopic picture is considered typical of that found in smears from persons infected with gonococci.
If the smears are properly prepared, a negative result in suspected acute cases is fairly conclusive evidence of the absence of gonococci. In chronic cases it is of only limited value and should be confirmed by further examinations.
It is especially difficult to demonstrate intracellular gonococci in chronic gonorrhea of the female. Too much reliance should not be placed on any single negative laboratory examination.

B. Complement-fixation tests:
Complement-fixation tests for gonococcal infection are of some value only in chronic conditions such as arthritis or endocarditis occurring as sequelae to possible gonococcal infections. A negative result is not conclusive. Complement-fixation tests are of no value in acute gonorrhea.

GRANULOMA INGUINALE

1. *Etiologic agent.* Donovan body, *Donovania granulomatis (Calymmatobacterium granulomatis; Klebsiella granulomatis).*

2. *Source of infection.* Discharges from lesions.

3. *Mode of transmission.* Presumably by direct contact of skin and mucous membranes during sexual intercourse with infected persons. The exact method of transmission is not known.

4. *Prevalence.* Widely prevalent in tropical and subtropical areas; endemic and recognized with increasing frequency in the United States.

5. *Current laboratory services.* Microscopic examination of scrapings from ulcers for Donovan bodies.

6. *Collection of specimens.* Take scrapings of material from ulcers and make smears on microscope slides contained in a "GC", "VI" or "MA" outfit. Mark history blank plainly "For Donovan bodies".

7. *Limitations of laboratory tests.* This disease is not to be confused with lymphogranuloma venereum (sometimes called lymphogranuloma inguinale). Laboratory findings are of value only to furnish presumptive evidence in support of clinical findings and history. Negative laboratory findings are not conclusive. Some prefer histologic examination of biopsy material to the use of smeared preparations. According to some authorities, the Donovan bodies are bacteria similar to Friedlander's bacillus; some consider these organisms to be secondary invaders.

HEPATITIS, INFECTIOUS

(ACUTE CATARRHAL JAUNDICE, EPIDEMIC JAUNDICE)

1. *Etiologic agent.* A specific virus or viruses, relatively resistant to heat.

2. *Source of infection.* Discharges from alimentary tract of infected persons and possibly also from the nose and mouth. Blood from infected persons.

3. *Mode of transmission.* The usual mode of transmission is not known. Several epidemics caused by contaminated water, food or milk, and by direct personal contact have been reported. The disease may be transmitted by transfusion of whole blood, blood serum or plasma from infected persons, and also by accidental contamination of syringes or needles with traces of blood from such persons.

4. *Prevalence.* Occurs sporadically and in epidemics; the latter are most commonly reported from institutions and from rural areas. General incidence in rural and urban areas is about the same. Most common among children and young adults, the incidence

declining with advancing years. The incidence tends to be highest in the autumn and early winter in the temperate zone. Outbreaks have been common in military forces during wars.

5. *Current laboratory services.* None available at Bureau of Laboratories except for exclusion of leptospiral infection.

6. *Collection of specimens.* See under "Leptospirosis".

7. *Limitations of laboratory tests.* At present the lack of facilities for specific diagnostic aids constitutes the main limitation. Until such time as facilities for the study of virus infections are more generally available, recognition of this disease must remain a clinical problem.

HEPATITIS, SERUM
(HOMOLOGOUS SERUM JAUNDICE)

1. *Etiologic agent.* A specific virus or viruses, relatively heat-resistant.

2. *Source of infection.* Blood or blood products from an infected person.

3. *Mode of transmission.* By parenteral (intravenous, intramuscular or subcutaneous) inoculation of infected blood, plasma or serum; or by administration of prophylactic or therapeutic agents from syringes and needles contaminated with traces of blood from infected persons.

4. *Prevalence.* Not determined; because of difficulty of distinguishing serum hepatitis from infectious hepatitis. Estimates of the incidence among recipients of pooled blood products have varied from around two per cent to as much as seven per cent. A high incidence has also been found in certain clinics where therapeutic parenteral injections are given.

5. *Current laboratory services.* None available except for exclusion of leptospirosis.

6. *Collection of specimens.* See under "Leptospirosis".

HISTOPLASMOSIS

1. *Etiologic agent. Histoplasma capsulatum.*

2. *Source of infection.* Unknown. In nature the etiologic agent has been recovered from humans and from dogs, cats, rodents, skunks and opossum, and it has been recovered from the soil.

3. *Mode of transmission.* Unknown.

4. *Prevalence.* The few proved cases have a world-wide distribution.

5. *Current laboratory services.* Cultures of material from sputum, skin lesions, blood or biopsies. Serological tests are available through arrangement with cooperating laboratories.

6. *Collection of specimens.* For sputum use "MI" outfit, marking history blank plainly "For fungi". For blood culture use "BC" outfit. For other materials use "FI" outfit following the directions enclosed. For serological test use "MI" outfit, collect 5-10 ml. of blood, and mark history blank plainly "For histoplasmosis".

7. *Limitations of laboratory tests.* Isolation of the organism constitutes evidence of infection. Serological tests are of limited value, especially when negative.

IMPETIGO CONTAGIOSA

1. *Etiologic agent.* Probably staphylococci or streptococci. Crater-like ulcers of the corium commonly show mixed infection with both beta hemolytic streptococci and staphylococci.

2. *Source of infection.* Lesions on the skin of an infected person; possibly discharges from the nose and throat.

3. *Mode of transmission.* Directly by contact with the moist discharges of the skin lesions, or indirectly by contact with articles recently soiled by those discharges. The infection may be readily inoculated from place to place on the patient's body by scratching.

4. *Prevalence.* Common among children, especially in warm weather. Occurs sporadically and also in epidemics in nurseries for infants, children's institutions and summer camps. Likely to spread rapidly where measures of personal hygiene are neglected and where skin lesions are frequent following scratching. In some countries the association with scabies and pediculosis is common.

5. *Current laboratory services.* Cultural identification of organisms present in suspected lesions.

6. *Collection of specimens.* Use sterile swab in "HS" outfit or collection of pus or exudate from lesions from which crusts have been removed. Remove plug from tube enclosed in outfit and plunge swab after taking material into the agar in the bottom of tube. Swab must be left in that position and the plug inserted around other end of swab in the tube. Exercise aseptic precautions throughout. Mark history blank plainly "For identification of organism".

7. *Limitation of laboratory tests.* Bacteria present in skin lesions may occur as secondary invaders. In this disease, the appearance of the lesions is a greater diagnostic aid than cultures as the latter yield evidence of distinctly inferior value.

INFLUENZA

1. *Etiologic agent.* Two distinct types of virus, designated as Type A (*Tarpeia alpha)* and Type B (*Tarpeia beta),* have been identified. The more widespread recent epidemics have been associated with influenza A and its variants. Influenza B has usually been found in smaller and more localized outbreaks. In some epidemics neither Type A nor Type B has been found. Several other subtypes have been identified.

2. *Source of infection.* Probably discharges from the mouth and nose of infected persons and articles freshly soiled by such discharges.

3. *Mode of transmission.* Believed to be by direct contact, by droplet infection, or by articles freshly soiled with discharges of the nose and throat of infected persons.

4. *Prevalence.* Variable in pandemics, local epidemics and as sporadic cases, often unrecognized by reason of indefinite clinical symptoms. In epidemics may affect up to 50 per cent of the population within 4-6 weeks. Occurs pandemically at irregular intervals.

5. *Current laboratory services.* Red cell agglutination inhibition test for types available will be made upon request after two blood specimens are received, one taken during the acute illness and one during convalescence (3-4 weeks later). Certain virus laboratories elsewhere are equipped to isolate the virus from nasopharyngeal washings but such facilities may only be used for epidemiological investigations in widespread outbreaks.

6. *Collection of specimens.* During acute phase, collect 5-10 ml. of blood in sterile bottle in "MI" outfit and mark history blank plainly "For influenza". Repeat 3-4 weeks later. Identify specimens (1) as "acute phase" and (2) "convalescent". Test on first specimen will not be made until second has been received.
Contact Bureau of Laboratories before collecting any other type of specimen. Instructions will be given providing it is feasible to enlist the services of a virus research laboratory.

7. *Limitations of laboratory tests.* A distinct rise in inhibition titer shown by the convalescent as compared with the acute phase specimen is of distinct diagnostic value. Demonstration of the virus is confirmatory of clinical observations and indicative of the type of virus present but is a difficult procedure not uniformly successful.

KALA AZAR (VISCERAL LEISHMANIASIS)

1. *Etiologic agent. Leishmania donovani.*

2. *Source of infection.* Man is the usual source of infection, but there is some suggestion that dogs may act as reservoir hosts; however, the relation between canine infection and human kala-azar is not yet settled.

3. *Mode of transmission.* Through bite of infected sandflies of the genus *Phlebotomus.* The fly is infected by sucking the peripheral blood or by ingesting the parasite that may be present in the skin of an infected person. In the stomach of the fly, leishmania forms develop into typical elongated leptomonas which spread forward to proventriculus and buccal cavity. The elongated metacyclic forms which are attached to the walls of the pharynx are injected by the sandfly in the act of biting.

4. *Prevalence.* Endemic in China (chiefly north of the Yang-tse); India (Bengal and Assam); Europe (Mediterranean area: Southern France, Italy, Sicily, Greece, Malta, Corsica, Aegean Islands, South Spain, Portugal, Turkey, Yugoslavia, Hungary); North Africa (Morocco, Algeria, Tunis); some areas in tropical Africa (Sudan, Ethiopia, Northern Kenya); South America (Argentina and Brazil); and also Transcaucasia, Russian Turkestan, South Manchuria, Palestine and Transjordania. In the Mediterranean areas the disease occurs among infants and young children. In Asiatic endemic areas the highest incidence is among children between the ages of five and fifteen years.

5. *Current laboratory services.*

   A. Microscopic examination of blood films.

   B. Complement-fixation tests on blood made through arrangements with cooperating laboratories.

6. *Collection of specimens.*

   A. Use "MA" outfit following directions enclosed as for malaria examination. Mark history blank plainly "For Leishmaniasis". Submit a series of 3-4 such specimens.

   B. Collect 5-10 ml. whole blood in "MI" outfit; Mark history blank plainly "For Leishmaniasis".

7. *Limitations of laboratory tests.* A single negative blood film has no significance; the organisms are rare in the peripheral blood. A negative serological test has no exclusion value.

KERATO-CONJUNCTIVITIS, INFECTIOUS (SUPERFICIAL PUNCTATE KERATITIS, NUMMULAR KERATITIS)

1. *Etiologic agent.* The virus of kerato-conjunctivitis.

2. *Source of infection.* Probably the discharge from the eye of an infected person or a carrier.

3. *Mode of transmission.* Apparently contact with an infected person or carrier or with articles freshly soiled with discharges of such person.

4. *Prevalence.* Occurs in epidemic form in warm climates, also among industrial employees in temperate climates involving a small percentage of the individuals in the groups affected.

5. *Current laboratory services.* None available at present at Bureau of Laboratories except cultures for exclusion of bacterial conjunctivitis. Certain virus laboratories have done experimental work which shows promise of future application.

6. *Collection of specimens.* Use "KL" outfit. Collect exudate from conjunctiva on one of swabs in envelope and immediately rotate swab gently but firmly over medium in one of the vials. If both eyes are affected use other swab and vial in outfit. Mark history blank plainly "For conjunctivitis", scratching out reference to examination for diphtheria; better still, use separate sheet of paper for pertinent information.

7. *Limitations of laboratory tests.* Certain pyogenic bacteria may be present in cultures as a result of contamination from adjacent skin. Failure to find a bacterial pathogen lends support to a diagnosis of infectious keratoconjunctivitis. (The Bureau of Laboratories may be able to arrange for special serological tests in a collaborating laboratory when outbreaks of this disease occur.)

LEISHMANIASIS, AMERICAN

(MUCOCUTANEOUS LEISHMANIASIS, ESPUNDIA, UTA, BUBOS)

1. *Etiologic agent. Leishmania brasiliensis.*

2. *Source of infection.* Infected persons.

3. *Mode of transmission.* Presumably through the bite of infected sand flies of the genus *Phlebotomus;* also by direct contact with infected individuals.

4. *Prevalence.* Reported from every country in South America except Chile and from Central America and Mexico. Most common among forest laborers. Both the cutaneous leishmaniasis due to *L. donovani* and the mucocutaneous leishmaniasis due to *L. brasiliensis* may be seen in the United States in persons coming from endemic areas.

5. *Current laboratory services.*

   A. Microscopic examination of scrapings from suspected lesions.

   B. Complement-fixation tests on blood made by cooperating laboratories by special arrangement.

6. *Collection of specimens.*

   A. Use "MA" outfit. Spread scrapings from suspected lesions on microscope slides and allow to dry. Mark history blank plainly "For *Leishmania*".

B. Collect 5-10 ml. of blood in sterile bottle in "MI" outfit. Mark history blank plainly for "Complement-fixation test for Leishmaniasis".

7. *Limitations of laboratory tests.* Identification of *Leishmania* in stained preparations is diagnostic of the disease when made by a specially trained observer. The complement fixation test is reliable when positive; negative reactions do not exclude infection.

LEPROSY (HANSEN'S DISEASE)

1. *Etiologic agent.* Leprosy bacillus, *Mycobacterium leprae.*

2. *Source of infection.* Discharges from lesions.

3. *Mode of transmission.* Intimate and usually prolonged contact with infected individuals. Some other as yet undetermined factors are apparently necessary.

4. *Prevalence.* Endemic in some Gulf coast areas of the United States, in Hawaii, Philippines and Puerto Rico. Prevalence practically confined to tropical and subtropical areas. Usually more frequent among adolescent and young adult males.

5. *Current laboratory services.* Microscopic examination of smears from cutaneous lesions and of swabbings from nasal septum. No other laboratory aids to diagnosis have proved practicable.

6. *Collection of specimens.* Smear scrapings or exudates from cutaneous lesions on microscope slides contained in "MA" outfit. Also submit smear of swabbings from nasal septum obtained by rubbing the septum briskly. Mark history blanks plainly "For leprosy".

7. *Limitations of laboratory tests.* Positive results indicate that acid-fast bacilli occurring in "lepra cells" or in packets characteristic of *Mycobacterium leprae* have been observed. The organism is otherwise morphologically identical with that of tuberculosis. Cultures and animal inoculations are not feasible. There is no practicable laboratory aid to diagnosis of those types of leprous infection not complicated by cutaneous lesions except the nasal swabbing technic.

LEPTOSPIROSIS (HEMORRHAGIC JAUNDICE, ICTEROHEMORRHAGIC SPIROCHETOSIS, WEIL'S DISEASE, CANICOLA FEVER, SWINEHERD'S DISEASE, MUD FEVERS)

1. *Etiologic agent.* A number of species of *Leptospira* including *L. icterohemorrhagiae* and *L. canicola.*

2. *Source of infection. Leptospira icterohemorrhagiae* is the cause of this common enzootic disease in wild rats. After infection they harbor these organisms in the kidney for long periods of time and excrete them in the urine. This is a common source of human infections. An analogous disease of dogs, known as "the yellows", is caused by *Leptospira canicola,* and in some localities gives rise to human cases of leptospiral jaundice. Other animals including cattle have occasionally been found to be infected with leptospira. *Leptospira pomona* has been incriminated in human infections in the United States. It is now recognized that many cases of leptospirosis show no jaundice and that symptoms may be diverse, including meningitis.

3. *Mode of transmission.* A majority of human infections result from contact of skin, abraded or not, or mucous membrane with water polluted with the urine of infected rats. Leptospiral meningitis has followed immersion in polluted water. Thus, the disease shows selection for such trades as fish dealers, abattoir workers, poultry dressers, sewer workers, agricultural workers, miners and veterinarians. Accordingly, it is largely a disease of male adults. Occasional infections result from handling dogs and other animals. The leptospira can penetrate uninjured skin.

4. *Prevalence.* The disease is present in rats over the entire world. Sporadic human cases have been reported throughout the United States.

5. *Current laboratory services.*
   A. Serological tests.
   B. Animal inoculation tests.

6. *Collection of specimens.*

   A. Collect blood for serological test (only after first ten days of illness) in sterile bottle in "MI" outfit. Mark plainly "For leptospirosis". Collect second specimen one week later.

   B. Collect blood (about 5 ml.) for animal inoculation (during first 3-5 days of illness only) in sterile bottle in "MI" outfit. Indicate duration of disease on history blank. Plainly mark history blank "For leptospira". Urine for animal inoculation should not be collected unless more than five days have elapsed from onset. Submit urine specimen in sterile bottle in "MI" outfit.

7. *Limitations of laboratory tests.* Failure to demonstrate a serological reaction is presumptive, but not necessarily conclusive, evidence

that jaundice observed clinically is due to other causes, including a virus. A rise in titer between the first and second specimen is diagnostic. *Leptospira* are sometimes difficult to demonstrate and artifacts in blood are sometimes mistaken for spiral organisms. Although direct examination of blood early in the disease by dark-field examination and by study of thick films is sometimes successful, animal inoculation tests on blood are more likely to be successful.

LYMPHOCYTIC CHORIOMENINGITIS

1. *Etiologic agent.* The virus of lymphocytic choriomeningitis (*Legio erebea*).

2. *Source of infection.* A reservoir of virus found in house mice (*Mus musculus musculus*).

3. *Mode of transmission.* The virus escapes from infected animals in mouth and nasal secretions, urine and feces. Transmission to man is probably through infected food or dust, possibly occasionally by arthropods. Not known to be communicable from man to man.

4. *Prevalence.* Rare but more common than the number of recognized cases indicates.

5. *Current laboratory services.*

   A. Complement-fixation tests on blood serum.

   B. Bacteriological examination of spinal fluid to eliminate bacterial meningitis.

   C. A limited number of tests to isolate virus from spinal fluid by arrangement with cooperating laboratories.

6. *Collection of specimens.*

   A. Collect two specimens of whole blood, 10-15 ml. each, in "MI" outfit, one specimen early in disease and one 3-4 weeks later. Mark history blanks "For lymphocytic choriomeningitis" or "For encephalitis".

   B. Spinal fluid for exclusion of bacterial meningitis may be submitted in "SF" outfit.

   C. Spinal fluid for isolation of the virus may be collected in "MI" outfit; it must be frozen immediately and kept frozen until examined. Contact Bureau of Laboratories before taking specimen.

7. *Limitations of laboratory tests.*

   A. Increase in titer of complement fixing antibodies in the blood during convalescence as compared with the blood titer during illness is demonstrable in some cases and is strong evidence of infection.

B. Negative findings tend to exclude bacterial etiology.

C. Isolation of the virus from spinal fluid is possible only in the first few days of the disease and provides conclusive diagnostic evidence but is not too practicable since specimens must be sent to a virus laboratory in frozen state. (Cell counts, preferably made locally, and total protein tests on spinal fluid may be helpful but are not specific.)

LYMPHOGRANULOMA VENEREUM* (CLIMATIC BUBO)

1. *Etiologic agent.* The virus of lymphogranuloma venereum (*Miyagawanella lymphogranulomatis)* immunologically related to the virus of psittacosis.

2. *Source of infection.* Discharges from lesions.

3. *Mode of transmission.* Direct contact by skin and mucous membranes, almost exclusively in sexual relations with infected persons.

4. *Prevalence.* A venereal infection not uncommon among Negroes in the poorer sections of American cities. Widely prevalent in the tropics and common among inmates and clients of brothels in seaports.

5. *Current laboratory services.* Complement fixation tests.

6. *Collection of specimens.* Collect about 5 ml. of blood, using aseptic precautions, and place in sterile bottle in "MI" outfit. Allow to clot firmly. Store in refrigerator if held for any length of time before transmitting to laboratory. Mark history blank plainly "For lymphogranuloma venereum".

7. *Limitations of laboratory tests.* Complement fixation findings correlate with Frei test findings in 70-80% of cases. Negative complement fixation tests do not conclusively rule out this infection. Positive findings are known to occur in the absence of this infection when patient has cirrhosis of the liver or an infection with certain related viruses (psittacosis, virus pneumonias). It has also been reported that early syphilis and granuloma inguinale may give some false reactions. Nevertheless, the test is a distinct aid in the confirmation of clinical findings.
(Intradermal (Frei) tests are also recommended.)

*Also known as Lymphopathia venereum and Lymphogranuloma inguinale.

MALARIA

1. *Etiologic agent.* The several species of micro-organisms: *Plasmodium vivax* (tertian); *P. malariae* (quartan); *P. falciparum* (estivo-autumnal or malignant subtertian); *P. ovale.*

2. *Source of infection.* The blood of an infected individual.

3. *Mode of transmission.* By bite of infected female anopheline mosquitoes. The mosquito is infected by biting an individual suffering from acute or chronic malaria. The parasite develops in the body of the mosquito for a variable period of time depending on the external temperature, under favorable conditions from 10 to 14 days (21 days for quartan), after which time the sporozoites appear in its salivary glands. The disease may be transmitted also by blood transfusion or by injecting whole human blood; also by common use of unsterilized hypodermic syringe (as by drug addicts). Mosquitoes of the genus *Anopheles* are the only known vectors.

4. *Prevalence.* Widespread in tropical and subtropical areas.

5. *Current laboratory services.* Identification of species of *Plasmodium* by microscopic examination of combined thick and thin blood films.

6. *Collection of specimens.* Preferably collect at least two specimens, one to be taken 12-24 hours after a chill and the other just before a chill is expected to recur. Each time collect both thick and thin blood films using slides in "MA" outfit, as follows:

   A. Thick film: Cleanse the ball of a finger or toe or the ear lobe with alcohol, allow to dry and puncture with a sterile lance or needle having a cutting edge. Avoid excessive bleeding but puncture deeply enough to produce a flow of blood with only slight pressure since bleeding produced by hard squeezing consists chiefly of serum which is unsatisfactory for reliable examination. The thick film should cover an area about the size of a dime. In judging the proper thickness, ordinary printing can just be read through the center of the wet film, which should be several layers of erythrocytes thick in the center with thinner edge. Collect blood on slide in one of the following ways.

      a. Touch under surface of slide about ¼ inch from end opposite the etched portion to the large, rotund drop of blood on the punctured skin and without losing contact with the drop or touching the skin move the slide in narrow circles until a smear of satisfactory thickness and size is made.

      b. Place 3-5 average drops of blood close together on the slide about ¼ inch from the end opposite the etched portion and immediately puddle these into one homogeneous drop of proper size using a needle or the corner of a clean slide.

B. Thin film: After preparing the thick film place a small drop of blood on the slide near the etched portion (not on it) opposite the thick film. Place the narrow edge of another slide held at an angle of about 45° across this drop which will instantly spread all along the line of contact between the slides. Then, without pressure push the inclined slide forward toward but not into the thick film, leaving behind a thin, smooth blood film.

Allow both thick and thin films to dry in air with slides on a flat surface. Do not apply heat. Do not replace in slide holder until fully dry. Protect from flies or other insects.

7. *Limitations of laboratory tests.* Light infections may be missed, particularly with the thin film only. A few parasites may be found in the thick film only in such cases. Because of the fact that *Plasmodium falciparum* (aestivo-autumnal parasite) disappears from the peripheral blood during the second 24 hours of the cycle, a series of examinations is more frequently necessary to detect this parasite than for detection of the other species. Proper identification of the parasites requires observation of the films by a specially trained microscopist.

MEASLES (RUBEOLA, MORBILLI)

1. *Etiologic agent.* The virus of measles (*Briareus morbillorum*).
2. *Source of infection.* Secretions of nose and throat of an infected individual.
3. *Mode of transmission.* By droplet spread directly from person to person; indirectly through articles freshly soiled with the secretions of the nose and throat of an infected individual. One of the most easily transmitted of the communicable diseases.
4. *Prevalence.* Universal. Probably 80 to 90 per cent of all persons surviving to the twentieth year of life have had an attack, and rarely does a person go through life without having had measles. Occurs most commonly in children. Endemic in large population units, with most cases in spring.
5. *Current laboratory services.* No practicable laboratory aid to diagnosis known to Bureau of Laboratories.

MENINGITIS, BACTERIAL INCLUDING MENINGOCOCCUS MENINGITIS (CEREBROSPINAL FEVER) AND MENINGOCOCCEMIA

1. *Etiologic agent.* These include: Meningococcus, or *Neisseria meningitidis* (*N. intracellularis*), of which four main serologic types or groups are recognized; *Hemophilus influenzae* (chiefly in children under three years of age); *Mycobacterium tuberculosis;* common pyogenic organisms; many others in sporadic cases.

2. *Source of infection.* Discharges from the nose and throat of patients or carriers, as the organisms are commonly carried in the nasopharynx. Meningococcus carrier prevalence of 25 per cent or higher may exist without the occurrence of cases. During epidemic periods more than half of a military organization may be healthy carriers of the strain of meningococci responsible for the epidemic.

3. *Mode of transmission.* By contact with infected persons, that is, sick persons or carriers. Indirect transmission may perhaps occur through contact with articles freshly soiled with discharges from the respiratory tract of infected persons.

4. *Prevalence.* Endemic and epidemic. There are no limits in geographic distribution. Sporadic cases occur throughout the year in both urban and rural areas with the greatest incidence during the winter and spring. The meningococcal disease exhibits high incidence at irregular intervals. The epidemic wave lasts usually two to three years.

5. *Current laboratory services.*

   A. Bacteriological examination of spinal fluid from cases of meningitis.

   B. Blood culture service in cases of meningococcemia.

   C. Complement-fixation tests for exclusion of certain viral infections.

6. *Collection of specimens.*

   A. Collect aseptically about 10 ml. spinal fluid in the sterile bottle supplied in the "SF" outfit. Check examinations desired on history blank. Rush to laboratory.

   B. Use "BC" outfit. Collect venous blood for culture aseptically in sterile syringe of 5-10 ml. capacity. Preferably replace used needle with another unused sterile one. Plunge needle through thin rubber top of stopper (previously swabbed with alcohol) and expel contents of syringe into medium in bottle. Rush to laboratory.

   C. Use "MI" outfit. Collect 10-20 ml. of blood during acute phase and again 3-4 weeks later. Mark history blank plainly "For encephalitis".

7. *Limitations of laboratory tests.* The laboratory procedures outlined above will detect infections other than meningococcic but animal inoculation tests on spinal fluid may be necessary if tuberculous meningitis is suspected. Cell counts (preferably made locally), total protein determinations and colloidal gold tests on spinal fluid may be valuable aids. *Neisseria meningitidis* (meningococcus), does not remain viable for long periods of time outside the body and it is therefore necessary to rush culture material to the laboratory as soon as possible. For this and other reasons, nasopharyngeal cultures for the detection of suspected carriers are

not feasible services for a central laboratory although these may be desirable to control an outbreak when facilities are available locally. The use of sulpha drugs to reduce the carrier rate in a closed population has met with some success. Complement-fixation tests for viral infections help to differentiate viral infections of the central nervous system from those of bacterial origin.

MONONUCLEOSIS, INFECTIOUS (GLANDULAR FEVER)

1. *Etiologic agent.* Unknown; possibly a virus.
2. *Source of infection.* Probably discharges from the nose and throat of infected persons.
3. *Mode of transmission.* Unknown.
4. *Prevalence.* Observed in many parts of the world and is probably much more prevalent and more widely distributed than indicated by reported incidence. Epidemics are most frequently recognized in schools and children's institutions; the recognized incidence is comparatively high among medical students, nurses, hospital personnel and among other groups having acccess to medical services where blood examinations are made routinely.
5. *Current laboratory services.* Serological examination of blood for heterophile agglutinins specific for infectious mononucleosis.
6. *Collection of specimens.* Collect 5-7 ml. venous blood aseptically and place in sterile bottle in "MI" outfit. Mark history blank plainly "For infectious mononucleosis".
7. *Limitations of laboratory tests.* Comparison of titers of serial specimens is helpful in cases that do not show conclusive serological evidence. Sheep red cell agglutinins may be present in the blood either naturally (native antibodies) or as a result of serum therapy (serum sickness) as well as in infectious mononucleosis. Since the simple agglutination test (Paul-Bunnell test) does not differentiate among these conditions, it is necessary to absorb the serum with guinea pig kidney before positive results of the test can be considered specific for infectious mononucleosis. This absorption removes all other heterophile antibodies. (Examination of a blood smear by a competent hematologist may provide evidence of the disease in very early cases or in those which do not develop the characteristic heterophile agglutinins.)

MUMPS (INFECTIOUS PAROTITIS) (INCLUDING MUMPS MENINGOENCEPHALITIS)

1. *Etiologic agent.* The virus of mumps (*Rabula inflans*).

2. *Source of infection.* Saliva of infected persons.

3. *Mode of transmission.* By droplet spread and direct contact with an infected person or with articles freshly soiled with the saliva of such persons.

4. *Prevalence.* This disease is less prevalent than the other common communicable diseases of childhood such as measles, whooping cough and chickenpox. Winter and spring are the seasons of greatest prevalence. Its occurrence is sporadic and epidemic except in large cities, where it is endemic. Outbreaks occur more frequently and are of a more serious character in aggregations of young people, especially military.

5. *Current laboratory services.* Complement-fixation tests for clinically atypical mumps infections.

6. *Collection of specimens.* Collect 5-10 ml. of blood in sterile bottle in "MI" outfit. Mark history blank plainly "For mumps". Collect first specimen early in disease and follow by a second specimen taken 3-4 weeks later.

7. *Limitations of laboratory tests.* Typical cases of mumps with parotid gland involvement seldom require laboratory confirmation. When mumps virus is the suspected cause of other types of illness including encephalitis, the complement-fixation test is a diagnostic aid of considerable value. Results are considered significant when the convalescent serum shows a rise in titer above the acute phase serum.

MYCOTIC INFECTIONS, MISCELLANEOUS*

1. *Etiologic agent.* Among others are *Candida albicans* (*Monilia albicans, Monilia candida*), *Blastomyces dermatitidis, Cryptococcus hominis* (*Torula histolytica*).

2. *Source of infection.* The organisms probably are widely distributed in nature but infection may occur from contact with such sources as infected animals.

3. *Mode of transmission.* By contact with infected materials or animals; trauma may be a contributing factor.

4. *Prevalence.* Relatively infrequent but worldwide in distribution.

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*Including Blastomycosis (Gilchrist's Disease), Paracoccidioidal Granuloma (Lutz-Splendore-De Almeida's Disease), Geotrichosis, Chromoblastomycosis (Chromomycosis, Verrucous Dermatitis), Cryptococcosis (Torulosis, Busse-Buschke's Disease), Candidiasis (Moniliasis, Thrush, Mycotic Vulvovaginitis, Bronchomycosis), Sporotrichosis, Maduromycosis (Madura Foot, Mycetoma), Aspergillosis, Rhinosporidiosis, Otomycosis, etc.

5. *Current laboratory services.* Microscopic and cultural examinations of body fluids, exudates, etc.

6. *Collection of specimens.* Use "FI" outfit. Collect body fluids, pus or other material from affected areas. Inoculate material on surface of medium in enclosed bottle; then make smears on enclosed microscope slides. For sputum preferably submit in "MI" container and mark history blank plainly "For fungi".

7. *Limitations of laboratory tests.* Cultural identification of known pathogenic types is desirable in all cases because of the widespread distribution of fungi in nature.

   (Intradermal tests are of value for some types but antigens are usually available only through research laboratories.)

ONCHOCERCIASIS

1. *Etiologic agent.* A nematode worm, *Onchocerca volvulus.*

2. *Source of infection.* The infected individual with microfilariae in the skin layers.

3. *Mode of transmission.* In Guatemala and Mexico, the bite of the following species of black flies: *Simulium callidum, S. metallicum* and *S. ochraceum.* In tropical Africa, *S. damosum* and *S. neavei.*

4. *Prevalence.* In the Western Hemisphere, limited regions of Guatemala, and the states of Chiapas and Oaxaca in Mexico. In Africa, Sierra Leone, the Gold Coast, Liberia, and the Belgian Congo up to the Sudan and Kenya.

5. *Current laboratory services.* Examination of biopsy material from skin, conjunctiva, or nodules.

6. *Collection of specimens.* Contact Bureau of Laboratories for instructions before taking specimens.

7. *Limitations of laboratory tests.* The presence of microfilariae in biopsies of skin or conjunctiva or of adult filariae in biopsied nodules constitutes a definitive diagnosis.

PARATYPHOID FEVER*

1. *Etiologic agent.* Paratyphoid bacillus A, B or C (*Salmonella paratyphi, Salmonella schottmuelleri, Salmonella hirschfeldii);* at times other *Salmonella.*

2. *Source of infection.* Feces and urine of patients or carriers. Temporary carriers may be numerous in an outbreak.

3. *Mode of transmission.* Transfer of micro-organisms in feces and urine through direct or indirect contact with patient or carrier.

*See also "SALMONELLOSIS".

Principal vehicles for indirect spread are contaminated food, water, milk, and shellfish, and, under some conditions, flies.

4. *Prevalence.* Frequency has fallen with that of typhoid fever. Occurring sporadically or in limited outbreaks due to contact or to contaminated food (milk, water). Probably more common than recognized due to the large number of unrecognized infections. Paratyphoid A fever much less common than B.

5. *Current laboratory services.*

   A. Blood cultures (1-7 days' duration of disease).

   B. Agglutination tests (after 10th day of illness). Cultures are made from the clots after the serum has been removed.

   C. Feces, urine and bile cultures for isolation and typing of organism.

6. *Collection of specimens.*

   A. Blood cultures: Use "BC" outfit. Collect aseptically venous blood for culture in sterile syringe of 5-10 ml. capacity. Preferably replace needle used with another unused sterile one. Plunge needle through thin rubber top of stopper (previously swabbed with alcohol) and expel contents of syringe into medium in bottle. Rush to laboratory.

   B. Agglutination tests and clot cultures: Collect 5-10 ml. venous blood aseptically in sterile bottle in "TY" outfit. Allow to clot firmly before sending to laboratory. A series of two or more specimens may be necessary to demonstrate increase in titer which usually reaches its height during the third week of illness.

   C. Feces: Use "FE" outfit. Using the sterile swab in the outfit, transfer a portion of the stool about the size of a sphere ½ inch in diameter to the specimen jar and emulsify it in the glycerol solution. If stools are liquid, transfer to the specimen jar with a clean spoon or other implement a quantity approximately 2 ml. in volume. Do not use spoon or other implement for more than one sampling unless carefully sterilized in the interim. Care should be taken that the stool is passed into a vessel that has been thoroughly disinfected or sterilized.

   Urine: Use "UR" outfit. Have patient void urine into previously sterilized vessel and place 10-15 ml. in sterile bottle provided in outfit.

   Bile: Collect this material ordinarily only when carrier has been hospitalized for release from carrier state according to the requirements of the Sanitary Code of Connecticut. Use duodenal tube and place 10-15 ml. of biliary drainage in sterile bottle in "FE" outfit. Be sure to check history blank to indicate specimen is bile.

7. *Limitations of laboratory tests.*

   A. For early diagnosis a blood culture is the best diagnostic aid. It should be taken during the first five days of illness, after which the organisms usually disappear from the blood stream. Isolation of a properly typed *Salmonella* establishes the cause of illness.

   B. Agglutinins do not appear in the blood until 10-14 days after onset. The titer reaches its height during the third week of illness. Agglutinins produced as a result of previous infection or of prophylactic vaccination may persist for an indefinite period of time. Therefore, the presence of agglutinins is not diagnostic of itself and must be considered in the light of clinical findings and of past history of the patient. Rises in titer shown by serial specimens are helpful. Since some patients may not develop agglutinins, negative results are only suggestive, not conclusive, of absence of this disease.
   Results of clot cultures have the same significance as those on blood cultures (See A above). Negative results are, however, likely to occur after the first five days of illness.

   C. Isolation of a properly typed *Salmonella* from the feces, urine or bile may mean the patient is a temporary or chronic carrier or that the organism isolated is the cause of the illness. Hence, the significance of positive results must be considered in the light of clinical findings. For release of cases and carriers, see the requirements of the Sanitary Code of Connecticut.

PEDICULOSIS (LOUSINESS)

1. *Infesting agent.* Head louse or body louse (*Pediculus humanus**), and crab louse (*Phthirus pubis*).

2. *Source of infestation.* Infected persons or their personal belongings, particularly body clothing.

3. *Mode of transmission.* Direct contact with an infested person and indirectly by contact with clothing and headgear of such persons.

4. *Prevalence.* Universal where there is neglect of washing of the person and the body clothing. The head louse is common in outbreaks among school children.

5. *Current laboratory services.* The diagnosis of this condition is not a laboratory function but identification of species of lice found, when there is some doubt of their identity, can be arranged through Bureau of Laboratories.

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*Includes both head and body lice, now regarded as two races of the same species.

6. *Collection of specimens.* Place lice in alcohol or 10% formalin in any convenient tightly stoppered vial.

7. *Limitations of laboratory tests.* Occasionally other lice or mites, such as chicken lice, may be mistaken for head or body lice. Examination under a low-power lens will bring out distinguishing characteristics.

PEMPHIGUS NEONATORUM (IMPETIGO OF THE NEWBORN)

1. *Etiologic agent.* Probably staphylococci which may be combined with streptococci.

2. *Source of infection.* Infected infants, attendants, or visitors.

3. *Mode of transmission.* By direct or indirect contact with infected persons or articles contaminated by them.

4. *Prevalence.* Occurs occasionally in nursery wards. Likely to spread rapidly.

5. *Current laboratory services.* Cultural identification of organisms present in suspected lesions.

6. *Collection of specimens.* Use sterile swab in "HS" outfit for collection of pus or exudate from lesions from which crusts have been removed. Remove plug from tube enclosed in outfit and plunge swab after taking material into the agar in the bottom of tube. Swab must be left in that position and the plug inserted around other end of swab in the tube. Exercise aseptic precautions throughout. Return outfit to laboratory by the quickest possible method. Delay in transit may result in false negative results. Mark history blank plainly "For identification of organism".

7. *Limitations of laboratory tests.* Bacteria present in skin lesions may occur as secondary invaders. In this disease, the appearance of the lesions is a greater diagnostic aid than cultures as the latter yield evidence of distinctly inferior value.

PERTUSSIS (WHOOPING COUGH)

1. *Etiologic agent.* Pertussis bacillus, *Hemophilus pertussis.*

2. *Source of infection.* Discharges from the laryngeal and bronchial mucous membranes of infected persons.

3. *Mode of transmission.* By direct contact with an infected person, or with articles freshly soiled with the discharges of such person.

4. *Prevalence.* Very prevalent, and a common disease among children everywhere regardless of race, climate or geographic location. Approximately only 15 per cent of the cases occur in children under two years of age, but 85 per cent of the deaths are in this age group. Seasonal incidence variable, but mortality higher usually in spring months in North America.

5. *Current laboratory services.* Cough plate cultures are feasible under certain conditions but are not routinely made at Bureau of Laboratories. Since this service cannot be extended to physicians on a statewide basis, only unusual circumstances justify examination of specimens.

6. *Collection of specimens.* Contact Bureau of Laboratories before submitting any specimens for this disease.

7. *Limitations of laboratory tests.* The causative organism, *Hemophilus pertussis,* will grow only upon specially prepared media and exposed cough plates must be placed under incubation immediately. A single negative culture is not conclusive.

PHLEBOTOMUS OR PAPPATACI FEVER (SANDFLY FEVER)

1. *Etiologic agent.* The virus of phlebotomus fever.

2. *Source of infection.* Blood of an infected person.

3. *Mode of transmission.* The vector is a small, hairy, blood-sucking midge, *Plebotomus papatasii,* which does most of its biting at night. It is possible that other species of *Phlebotomus* may also carry the virus.

4. *Prevalence.* Occurs only in those parts of Europe, Africa and Asia where the vector exists. A disease of subtropical and tropical areas where there are long periods of hot, dry weather, but in general is found in a belt extending around the Mediterranean Sea eastward into Burma and China. Seasonal, occurring between April and October, and prone to appear as a disease of troops from nonendemic areas who enter an endemic zone during the spring and summer.

5. *Current laboratory services.* No laboratory aid to diagnosis is known to the Bureau of Laboratories.

PLAGUE

1. *Etiologic agent.* Plague bacillus, *Pasteurella pestis.*

2. *Source of infection.* Infected rodents and patients with pneumonic plague. The primary or indigenous source of the disease is the plague of wild rodents, an animal reservoir that includes the ground squirrel (*Citellus beecheyi),* pack rats (*Neotoma)* and harvest mice (*Microtus)* of the United States and various species of wild rodents in South America, South Africa, Southeast Russia, eastern Asia, especially Mongolia and Manchuria, and other parts of the world. Infection may reach man from these sources, or more often through the medium of the domestic rat.

3. *Mode of transmission.* By droplet infection and directly in the pneumonic form. Bubonic plague is generally transmitted from

rats to man by the bites of fleas (*Xenopsylla cheopis* and certain other species), also by fleas from other rodents. Accidental infections, commonly of laboratory origin, arise through inoculation or inhalation of contaminated materials.

4. *Prevalence.* Rare in North America and insular possessions of the United States, usually occurring as sporadic cases from exposure to infected wild rodents west of the Mississippi. Focally distributed in various parts of the world.

5. *Current laboratory services.* Cultural examination of material aspirated from buboes, or of sputum in pneumonic type, can be made through special arrangements with Bureau of Laboratories.

6. *Collection of specimens.* Contact Bureau of Laboratories before collecting any specimens. Special instructions will be given. There is a federal prohibition against admission of plague specimens to the mails.

7. *Limitations of laboratory tests.* For precise identification of the causative organism, *Pasteurella pestis,* animal inoculations and serological studies are required.

PNEUMONIA

A. Pneumococcal — Acute Lobar Pneumonia

1. *Etiologic agent.* Pneumococci (*Diplococcus pneumoniae)* Types I to XXXII inclusive account for about 95 per cent of the cases, the remaining are due to the more rarely recognized types.

2. *Source of infection.* Probably discharges from the mouth and nose of infected persons. The pneumococcus is widely distributed in the upper respiratory tract discharges among healthy members of most communities.

3. *Mode of transmission.* By direct contact with infected person, or with articles freshly soiled with the discharges of the nose and throat of such person, and possibly from minute suspended particles containing the etiologic agent.

4. *Prevalence.* Common, and affecting at one time or other, between adolescence and old age, a large proportion of the population. No race or color and neither sex is exempt from likelihood of having this disease. Occurs in all climates and seasons, but most often in winter and spring and in regions where cold, windy, changeable and inclement weather prevails. Epidemic outbreaks of this disease occur in institutions, particularly those for adults, and in barracks.

5. *Current laboratory services.*

   A. Isolation and typing of organism.

   B. Blood cultures.

C. Cultural examination and typing of pneumococci isolated from body fluids or discharges when such complications or sequelae as otitis media or meningitis occur.

6. *Collection of specimens.*

A. Sputum for typing: Use "PN" outfit. Collect sputum which has been coughed up from the lungs. Avoid inclusion of saliva and superficial mucous secretions. Particularly include portions of sputum that are blood tinged. Place sputum in sterile bottle in outfit. On young children from whom sputum is difficult to obtain, material coughed up from the lungs may be obtained directly on a good sized, sterile swab held to the back of the mouth when coughing occurs or is induced. The handle of the swab may be broken off and the swab placed in the sterile bottle provided in the outfit.

B. Blood cultures: Use "BC" outfit. Collect aseptically venous blood for culture in sterile syringe of 5-10 ml. capacity. Preferably replace needle used with another unused sterile one. Plunge needle through thin rubber stopper (previously swabbed with alcohol) and expel contents of syringe into medium in bottle. Rush to laboratory.

C. Use "MI" outfit for body fluids; use "HS" outfit for pus or other discharges. Use aseptic technique. Mark history blank "For type of organism". Rush to laboratory.

7. *Limitations of laboratory tests.*

A. Sputum typing: Provided sputum is obtained from the deeper air passages, a negative specimen from individuals not treated with antibiotics or sulfa drugs is presumptive evidence of absence of pneumococcic infection.
The type of pneumococcus found in the sputum is considered of diagnostic significance in the absence of blood culture findings when symptoms are those of lobar pneumonia. Blood cultures should be taken to detect bacteremia and to follow results of treatment.
Other organisms such as *Klebsiella pneumoniae* (Friedlander's bacillus) and pyogenic cocci may produce pneumonia. *Mycobacterium tuberculosis* (tubercle bacillus) is also reported when found.

B. Blood cultures: A pneumococcus type isolated from blood culture is considered proof of etiological significance. Bacteremia may not occur except in severe cases.

C. Cultures of body fluids and discharges: The type of organism reported may be considered of significance unless introduced in the collection of the specimen.

B. Bacterial Pneumonia, other than Pneumococcal

1. *Etiologic agent.* Various pathogenic bacteria of the mouth, nose and throat, as streptococci, staphylococci, *Klebsiella pneumoniae* and *Hemophilus influenzae.*

2. *Source of infection.* Probably discharges from the mouth and nose of an infected person.

3. *Mode of transmission.* By direct contact with infected person or with articles freshly soiled with discharges of nose or throat of such person.

4. *Prevalence.* Common only during prevalence of epidemic influenza or of other respiratory infections.

5. *Current laboratory services.* Cultural identification of organisms in sputum.

6. *Collection of specimens.* Use "PN" outfit. Collect sputum which has been coughed up from the lungs. Avoid inclusion of saliva and superficial mucous secretions. Particularly include portions of sputum that are blood tinged. Place sputum in sterile bottle in outfit. On young children from whom sputum is difficult to obtain, material coughed up from the lungs may be obtained directly on a good sized, sterile swab held to the back of the mouth when coughing occurs or is induced. The handle of the swab may be broken off and the swab placed in a sterile bottle provided in the outfit.

7. *Limitations of laboratory tests.* Organisms other than the pneumococcus most likely to be of significance when found in pneumonic sputum are: *Klebsiella pneumoniae* (Friedlander's bacillus), beta hemolytic streptococci and sometimes *Hemophilus influenzae* or the pyogenic cocci. The presence of these organisms in sputum constitutes contributory evidence only and is not diagnostic.

C. Primary Atypical Pneumonia (Virus Pneumonia)

1. *Etiologic agent.* The causative agent of the majority of atypical pneumonias is probably a virus. See also "Psittacosis" and "Q Fever".

2. *Source of infection.* Probably discharges from the mouth and nose of infected persons or articles freshly soiled with such discharges.

3. *Mode of transmission.* By direct contact with infected person or with articles freshly soiled with discharges of nose and throat of such person. Mild, unrecognized infections may play a role in the spread of the disease.

4. *Prevalence.* Occurs endemically and in epidemics at all seasons. Incidence is variable. In outbreaks in army camps attack rates of from 1 to 6 per cent of the troops per year have been reported. Similar attack rates are reported in civilian hospitals and institu-

tions. Occurs in both sexes and at all ages, but is more frequent in adolescents and young adults.

5. *Current laboratory services.*

   A. For exclusion of bacterial agents, see preceding sections on "Pneumococcal" and "Bacterial Pneumonia".

   B. Serological tests on blood.

6. *Collection of specimens.*

   A. For examinations to aid in exclusion of bacterial pathogens, use "PN" outfit. Collect sputum which has been coughed up from the lungs. Avoid inclusion of saliva and superficial mucous secretions. Particularly include portions of sputum that are blood tinged. Place sputum in sterile bottle in outfit. On young children from whom sputum is difficult to obtain, material coughed up from the lungs may be obtained directly on a good sized sterile swab held to the back of the mouth when coughing occurs or is induced. The handle of the swab may be broken off and the swab placed in a sterile bottle provided in the outfit.

   B. Collect 5-10 ml. blood in sterile bottle in "MI" outfit. Mark history blank plainly "For virus pneumonia". Collect a second specimen during convalescence.

7. *Limitations of laboratory tests.*

   A. Results of examinations for the exclusion of bacterial pathogens are of indirect value only in the establishment of a diagnosis of primary atypical pneumonia as defined above.

   B. The demonstration of cold agglutinins (autohemagglutinins) is strong evidence for an atypical pneumonia presumably caused by a virus. The demonstration of a rising titer in serological tests for psittacosis or Q fever is evidence for the causative role of the psittacosis group of viruses or of *Coxiella burneti* respectively.

POLIOMYELITIS (INFANTILE PARALYSIS)

1. *Etiologic agent.* The virus of poliomyelitis (*Legio debilitans*). Several (at least 3) immunologically distinct types have been identified.

2. *Source of infection.* Pharyngeal (nose and throat) discharges and feces of infected persons, frequently those not suffering from a clinically recognized attack of the disease.

3. *Mode of transmission.* Close association with infected persons accounts for a large proportion of cases. Outbreaks attributable to milk have been rare and limited. Flies have been found to be

contaminated with the virus but there has been no reliable evidence of spread by insects, water, food or sewage.

4. *Prevalence.* Infection is prevalent throughout the world. Paralytic cases have been apparently more frequent in the temperate zones. Occurs both sporadically and in epidemics at irregular intervals, with the highest incidence in summer and fall. In the United States an annual incidence of 10 paralytic cases per 100,000 population is usual but there is a wide variation in incidence from year to year and region to region. Children from one to 16 years of age are more frequently attacked than adults. In several countries, including the United States, older children and young adults constitute a higher proportion of reported cases than formerly. Even during epidemics the incidence of paralytic cases has rarely exceeded 100 per 100,000 population.

5. *Current laboratory services.* It is recommended that complement-fixation tests for mumps be made on each suspected case in Connecticut, once during the early stage and again 3-4 weeks later. No specific aids available at Bureau of Laboratories. Routine examinations of spinal fluid for total protein and colloidal gold test are made when requested.

6. *Collection of specimens.* See under "Mumps" for collection of blood as recommended above. Spinal fluid for routine examination may be submitted in the "SF" outfit. Collect aseptically about 10 ml. of fluid in the sterile bottle in the outfit.

7. *Limitations of laboratory tests.* Many suspected abortive cases may be shown by serologic test to be mumps meningoencephalitis (or other viral infection other than poliomyelitis). Cell counts of spinal fluid, preferably made locally, may aid in making an early diagnosis. Total protein and colloidal gold reactions provide indirect information of limited value. Virus neutralization tests are not made for diagnostic purposes, even by research laboratories, because of difficulty of interpretation.

PROTOZOAN INFECTIONS, MISCELLANEOUS*

1. *Etiologic agent.* Among others, *Balantidium coli, Giardia lamblia, Chilomastix mesnili, Trichomonas vaginalis, Isospora hominis.*

2. *Source of infection.* Infected humans.

3. *Mode of transmission.* Direct contact or by articles or food soiled by infected persons. Flies may be an intermediary.

4. *Prevalence.* Widespread, usually sporadic but may occur in outbreak form especially in institutions.

*The infection, usually meningoencephalitis, with *Toxoplasma*, known as Toxoplasmosis, is a member of this group. Blood tests for toxoplasmosis are available through collaborating laboratories; contact Bureau of Laboratories before submitting specimen.

5. *Current laboratory services.* Microscopic examinations of excreta or other material for type of organism.

6. *Collection of specimens.* Feces or urine may be submitted in bottles in "PD" outfit. Vaginal smears for *Trichomonas vaginalis* may be submitted on microscope slides in "GC" outfit; examination for these parasites is made routinely on specimens for gonorrhea.

7. *Limitations of laboratory tests. Balantidium coli* is pathogenic. Species of *Giardia, Chilomastix* and *Trichomonas* are of doubtful pathogenicity except that the last may cause vaginitis. *Isospora hominis* is a cause of coccidiosis in man (not known in Connecticut).

PSITTACOSIS*

1. *Etiologic agent.* The virus of psittacosis (*Miyagawanella psittacii*) a specific virus related to the causal agent of lymphogranuloma venereum.

2. *Source of infection.* Parrots, parakeets, love birds, canaries, pigeons and other birds. Birds which are apparently well occasionally transmit the infection.

3. *Mode of transmission.* Contact with infected birds or their recent surroundings. Occasionally through a human case.

4. *Prevalence.* Usually in sudden house outbreaks among persons exposed to sick birds. Deaths mainly confined to persons over 30 years of age. Milder cases not infrequent from slight exposure to pigeons or other birds not necessarily sick.

5. *Current laboratory services.*

   A. Complement-fixation tests on blood.

   B. Tests for virus in sputum and other special examinations can be arranged through cooperating virus laboratories.

6. *Collection of specimens.*

   A. Collect 10-15 ml. of blood in sterile bottle in "MI" outfit. Mark history blank plainly "For psittacosis". Repeat after 3-4 weeks.

   B. Contact Bureau of Laboratories before submitting any specimens. Special instructions will be given.

7. *Limitations of laboratory tests.*

   A. Positive complement-fixation tests with the virus or a related one (lymphogranuloma venereum) are of significance when a rise in titer is found in a convalescent specimen as compared with an acute phase specimen.

*Includes the closely related "Ornithosis".

B. Identification of the virus by animal inoculation tests is of diagnostic significance. Development of neutralizing antibodies in the blood during convalescence is strong presumptive evidence of infection.

Q FEVER

1. *Etiologic agent. Coxiella burneti (Rickettsia burneti).*

2. *Source of infection.* Cows, sheep, goats, ticks and bandicoots appear to be natural reservoirs. The etiologic agent is abundant in the mammary gland and milk of infected cows; it has also been found in the milk of sheep and goats.

3. *Mode of transmission.* Not well understood. Occupational exposure appears to be important. Workers in stockyards, meat packing plants, dairies and diagnostic and research laboratories have accounted for a large proportion of the cases reported. Milk from infected cows appears to be an important means of spread in southern California. Commercial pasteurization appears to reduce the hazard from the use of milk but *C. burneti* have been demonstrated in commercially pasteurized milk. A number of species of ticks have been found naturally infected in the United States, Australia and Algeria, but their importance in transmission of the disease has not been demonstrated.

4. *Prevalence.* The disease has been reported from Australia, the United States, Panama, the Mediterranean basin, Switzerland, France and Portugal. In the United States, outbreaks, in addition to those among laboratory workers, have occurred among abattoir employees in Texas and Illinois. Endemic foci of the disease are known to exist in northern and southern California. Cases have been found also in Arizona, Minnesota, Montana, Pennsylvania and New York.

5. *Current laboratory services.*

   A. Complement-fixation tests on blood serum.

   B. Through cooperating laboratories, recovery of the organism in specimens from the patient or in suspected vehicles of infection can be attempted when circumstances warrant.

6. *Collection of specimens.*

   A. *Use "MI" outfit.* Collect 10-20 ml. blood during acute stage; repeat 3-4 weeks later. Mark history blank plainly "For Q fever" or "For Rickettsial infections".

   B. Contact Bureau of Laboratories before submitting any specimens.

7. *Limitations of laboratory tests.* A rise in titer of complement fixing antibodies in the second specimen as compared with the

first constitutes laboratory confirmation of clinical findings. Isolation of the infecting agent is a procedure which is hazardous to laboratory personnel; hence it should be attempted only in laboratories with special facilities only when other measures will not suffice.

RABIES

1. *Etiologic agent.* The virus of rabies (*Formido inexorabilis*).

2. *Source of infection.* Infected animals, chiefly dogs; vampire bats in limited areas.

3. *Mode of transmission.* Almost always bites by a rabid animal, on rare occasions through contact of such animal's saliva with scratch or other break in a person's skin. Milk or meat of infected animals, such as cows, is not dangerous for human use.

4. *Prevalence.* Incidence in man is low. Occurs on all continents except Australia. More prevalent among dogs and sometimes in wild carnivorous animals. Seasonal prevalence when observed is not related to temperature. Rabies occurs in vampire bats in Trinidad, certain areas of Brazil, Venezuela and Mexico. The virus isolated from the vampire bat is closely related to known strains of rabies virus from dogs and other animals.

5. *Current laboratory services.*

   A. **Animal rabies:** Microscopic examination of brain tissue for Negri bodies and animal inoculation tests for confirmation of negative results.

   B. **Human rabies:** Microscopic examination of brain tissue removed at autopsy and confirmatory animal inoculation tests.

6. *Collection of specimens.*

   A. **Animal heads:** Do not kill suspected animals unnecessarily. When clinical symptoms of the animal are suspicious begin antirabic treatment of persons bitten or otherwise exposed without waiting for a laboratory test. Keep suspected animals under observation according to instructions from your local director of health. A rabid animal does not recover; the disease is progressive and fatal. When suspected animal has died or has had to be killed, remove head and place in a metal container with a tightly fitting metal top. Pack this in ice before submission to the laboratory.

   B. **Human brains:** When permission for autopsy has been secured, place brain in suitable watertight container, pack in ice and rush it to the laboratory; do not place brain in fixative.

7. *Limitations of laboratory tests.* The finding of typical Negri bodies in nerve cells of the brain is considered diagnostic of rabies. Animal inoculation tests complete the evidence.

Brain tissue may not contain Negri bodies in detectable numbers early in the disease. Negri bodies may not be found if those portions of the brain where they are usually found have been destroyed by gunshot or violent blow. Decomposed brains are unreliable for examination and frequently unsuitable for animal inoculation tests.

RAT-BITE FEVER*

1. *Etiologic agent.* Either of two distinct organisms, *Streptobacillus moniliformis (Haverhillia multiformis, Streptothrix muris rattis, Actinoncyces muris)* or *Spirillum minus (Spirocheta morsusmuris).*

2. *Source of infection.* Usually bite of the rat, rarely of other rodents (squirrel, weasel). Sporadic cases without reference to bite have been recorded.

3. *Mode of transmission.* During the bite, animal blood escapes from the injured or diseased buccal mucosa into the wound, or the conjunctival secretion of the rat may contaminate the wound. Blood from an animal in the laboratory may infect man. Localized epidemics may occur from milk or milk products (Haverhill fever).

   The means of contamination is not known, whether through infection of cows by rat bite, or direct contamination of milk by rats.

4. *Prevalence.* Distribution is world-wide. Rare in North and South America and in most European countries.

5. *Current laboratory services.*

   A. Cultures of blood or of exudate from primary lesion for demonstration of the causative organism.

   B. Agglutination tests on blood for infection with *Streptobacillus moniliformis* are made through arrangements with cooperating laboratories.

   C. Animal inoculation tests when *Spirillum minus* infection is suspected.

6. *Collection of specimens.*

   A. Use "BC" outfit for blood culture; "HS" outfit for exudate from lesion.

   B and C. At height of fever collect 5-10 ml. of venous blood in sterile bottle in "MI" outfit. Mark history blank plainly "for rat-bite fever". Collect second specimen for agglutination test 10-14 days later.

7. *Limitations of laboratory tests.* Isolation of the organism is confirmatory of the clinical syndrome; agglutination tests have value only when reaction is strong.

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*Includes infection either with *Streptobacillus moniliformis* (Haverhill fever) and with *Spirillum minus (Sodoku).*

RELAPSING FEVER

A. Louse-Borne

1. *Etiologic agent.* A spirochete, *Borrelia recurrentis.*

2. *Source of infection.* The natural reservoir of infection is unknown. After biting an infected human being lice *(Pediculus humanus)* become infective in 6-8 days and remain so for life (30 or 40 days). Hereditary transmission in lice through the egg to the larval form is reported but has rarely been observed.

3. *Mode of transmission.* Transmission to man is effected by crushing an infected louse into the bite-wound or into an abrasion on the skin, or by rubbing louse feces or coxal fluid onto an abrasion of the skin. The louse becomes infective through its feces (not its bite) 8 days after it has sucked blood from an infected person.

4. *Prevalence.* The disease is prevalent among primitive people who are louse infested. It is found in limited localities in Europe, Asia, North and South Africa and Central America. For more than a quarter of a century louse-borne relapsing fever has not been observed in the United States.

5. *Current laboratory services.*

   A. Microscopic examination of thick blood films.

   B. Dark-field examinations and animal inoculation tests on blood specimens.

6. *Collection of specimens.*

   A. Blood films: During febrile paroxysm make thick blood film. (See directions under "MALARIA".) Allow to dry in air without application of heat before sending to laboratory.

   B. Whole Blood: During febrile paroxysm collect 15 ml. of venous blood aseptically and place in sterile bottle in "MI" outfit. Mark history blank plainly "For relapsing fever".

7. *Limitations of laboratory tests.* It is not possible to distinguish between species of *Borrelia* by ordinary laboratory examinations. Presence of any *Borrelia* in the blood is, however, confirmatory of the clinical syndrome of relapsing fever. Specimens taken during afebrile periods are likely to yield negative results.

B. Tick-Borne

1. *Etiologic agent.* A spirochete, *Borrelia duttoni.*

2. *Source of infection.* Primarily an infection of wild rodents, transmitted by the genus of ticks *Ornithodorus.* In the United States *O. turicata* is a known vector in Texas and Kansas; *O. hermsi* in California, Colorado and Idaho. *O. talaje* is a vector in Panama, Central and South America, while *O. moubata* is the vector in tropical Africa. Transovarian infection occurs in ticks.

3. *Mode of transmission.* Man is accidentally infected by a tick bite.

4. *Prevalence.* Widespread through tropical Africa. Foci have been observed in Spain, North Africa, Arabia, Iran, India and other parts of Central Asia as well as in North and South America. In the United States human cases of tick-borne relapsing fever have been found to originate in limited localities of 13 states — Arizona, California, Colorado, Idaho, Kansas, Montana, Nevada, New Mexico, Oklahoma, Oregon, Texas, Utah and Washington.

5. *Current laboratory services.*

   A. Microscopic examination of thick blood films.

   B. Dark-field examinations and animal inoculation tests on blood specimens.

6. *Collection of specimens.*

   A. Blood films: During febrile paroxysm make thick blood film. (See directions under "MALARIA".) Allow to dry in air without application of heat before sending to laboratory.

   B. Whole blood: During febrile paroxysm collect 15 ml. venous blood aseptically and place in sterile bottle in "MI" outfit. Mark history blank plainly "For relapsing fever".

7. *Limitations of laboratory tests.* It is not possible to distinguish between species of *Borrelia* by ordinary laboratory examinations. Presence of any *Borrelia* in the blood is, however, confirmatory of the clinical syndrome of relapsing fever. Specimens taken during afebrile periods are likely to yield negative results.

RHEUMATIC FEVER (ACUTE RHEUMATIC FEVER, ACUTE ARTICULAR RHEUMATISM)

1. *Etiologic agent.* Unknown. Attacks are usually precipitated by group A streptococcal respiratory infections. Thus an etiologic relationship is suspected but the mechanism is unknown. No specific infecting organism has been found in rheumatic fever lesions. The lesions resemble sensitization phenomena.

2. *Source of infection.* Unknown (See above).

3. *Mode of transmission.* Unknown.

4. *Prevalence.* There is a close parallelism between the prevalence of rheumatic fever and the prevalence of recognized streptococcal respiratory infections. In the United States rheumatic fever is most prevalent in the Rocky Mountain region and in the New England and North and Central Atlantic states. The lowest incidence of rheumatic fever occurs in the south and southwest. The curve of seasonal incidence follows that of streptococcal infections, but in the United States it reaches its peak during the spring months and its low point during the summer and early autumn.

5. *Current laboratory services.* No specific laboratory tests available.

RICKETTSIALPOX

1. *Etiologic agent.* *Rickettsia akari.*

2. *Source of infection.* Infected house mice (*Mus musculus musculus*).

3. *Mode of transmission.* The agent is transmitted from mouse to mouse and probably from mouse to man by a rodent mite (*Allodermanyssus sanguineus*).

4. *Prevalence.* The only cases reported up to this time (August, 1950) have been from New York City. Occurrence in other areas is probable.

5. *Current laboratory services.* Complement-fixation tests. Weil-Felix tests not recommended for this infection.

6. *Collection of specimens.* Use "MI" outfit. Collect 10-20 ml. of blood during early stage and repeat about 3 weeks later. Mark history blank plainly "For rickettsial infections".

7. *Limitations of laboratory tests.* A rise in titer of complement fixing antibodies in the second specimen as compared with the first is considered diagnostic of rickettsial infection; however, the rickettsialpox antigen reacts broadly in infections with other *Rickettsia* and, consequently, it is desirable to have tests for other rickettsial diseases for comparison.

RINGWORM (DERMATOPHYTOSIS, EPIDERMOPHYTOSIS)

A. Ringworm of the Scalp* (Tinea Capitis)

1. *Etiologic agent.* *Microsporon audouini, M. canis* (*lanosum* or *felineum*) and other species of fungi cause sporadic tinea capitis.

2. *Source of infection.* Lesions on scalps of infected persons; articles of clothing, especially hats and caps containing the fungus or its spores, or infected hairs or scales shed by individuals. In the case of infection with *M. canis* or other animal fungi, contact with lesions or hair shed by young cats or dogs affected with ringworm.

3. *Mode of transmission.* Directly from person to person by contact with lesions of infected persons (or in the case of animal fungi, with infected animals). Possibly indirectly by articles of wearing apparel or by surfaces contaminated with scales or hairs from lesions. Transmission occurs in the home and in schools, especially during games in which personal contact is close, and from barbers' instruments.

4. *Prevalence.* Widespread, with epidemics especially among school children and in institutions for children.

5. *Current laboratory services.* Microscopic and cultural examinations of hairs and skin scales from affected areas.

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*Also known as Trichophytosis capitis, Tinea tonsurans. See also "Favus".

6. *Collection of specimens.* Use "FI" outfit. With the aid of a Wood's ultraviolet lamp if necessary to locate isolated areas, collect hairs for microscopic and cultural examination as directed therein.

7. *Limitations of laboratory tests.* Cultural identification of fungi observed microscopically is desirable.

B. Ringworm of the Body* (including groin and feet)

1. *Etiologic agent.* Several species of fungi pathogenic to the skin.

2. *Source of infection.* Lesions on bodies of infected persons, articles of clothing carrying the fungus or its spores.

3. *Mode of transmission.* Directly by skin-to-skin contact with lesions of infected persons; possibly indirectly by articles of wearing apparel or towels or surfaces contaminated with scales or hair from such lesions.

4. *Prevalence.* Widespread, varying with conditions of crowding, and during periods of warm weather.

5. *Current laboratory services.* Microscopic and cultural examinations of skin scales from the edge of lesions.

6. *Collection of specimens.* Secondary allergenic lesions ("ids") may be sterile. A probable primary focus should be selected. Use "FI" outfit; collect skin scales from the edge of lesions as directed therein.

7. *Limitations of laboratory tests.* Cultural identification of fungi observed microscopically is desirable.

ROCKY MOUNTAIN SPOTTED FEVER (TICK-BORNE)

1. *Etiologic agent. Rickettsia rickettsi (Dermacentroxenus rickettsi).*

2. *Source of infection.* Infected ticks. In the eastern and southern United States the common vector is the dog tick, *Dermacentor variabilis;* in the northwestern United States, it is the wood tick, *Dermacentor andersoni;* in the southwestern United States it may occasionally be the lone star tick, *Amblyomma americanum.* In Brazil *Amblyomma cajennese* is the common vector. The rabbit tick (*Haemaphysalis leporis palustris)* has been found naturally infected, but this species does not bite man. The infection is passed from generation to generation in ticks and is probably maintained by infected and non-infected larvae feeding upon susceptible wild rodents.

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*Includes Tinea pedis (Epidermatophytosis, athlete's foot), Tinea unguium (Onychomycosis, Trichophyton unguium), Tinea cruris (Eczema marginatum, jockey itch, Dhobie itch, gym itch), Tinea corporis (Tinea circinata, Tinea glabrosa, Trichophyton corporis), Tinea imbricata, Tinea barbae (Tinea sycosis, barber's itch) and allied infections such as Piedra (Tinea nodosa), Trichomycosis axillaris, Tinea versicolor (Pityriasis versicolor, Tinea flava) and Erythrasma.

3. *Mode of transmission.* Bite of tick or contact with tick material such as its blood or feces on the unbroken skin.

4. *Prevalence.* Known to occur throughout North America, and in several areas of South America. The season of occurrence is predominantly in the spring and early summer, corresponding to the time of appearance of adult ticks.

5. *Current laboratory services.* Complement-fixation tests for rickettsial diseases.

6. *Collection of specimens.* Use "MI" outfit. Collect 10-20 ml. of venous blood during acute phase and repeat about 3 weeks later. Mark history blank plainly "For rickettsial diseases".

7. *Limitations of laboratory tests.* Positive complement-fixation tests are confirmatory of clinical findings and usually indicate the type of infection present. The reaction is usually negative until the 14th day after onset. Preferably demonstrate a rise in titer between the early specimen and one taken 3-4 weeks later.

RUBELLA (GERMAN MEASLES)

1. *Etiologic agent.* The virus of rubella.

2. *Source of infection.* Secretions of the mouth and possibly the nose.

3. *Mode of transmission.* By direct contact with the patient or with articles freshly soiled with the discharges from the nose or throat of the patient.

4. *Prevalence.* Epidemic in expression, occurring mostly in childhood, but more in adults than is the case with measles; more prevalent in winter and spring than at other seasons.

5. *Current laboratory services.* No practicable diagnostic laboratory aid known to the Bureau of Laboratories.

SALMONELLOSIS

1. *Etiologic agent.* A member of the genus *Salmonella.* Among the more common causes of infections in man in the United States are *Salmonella typhimurium, Salmonella choleraesuis, Salmonella newport, Salmonella oranienberg* and *Salmonella montevideo.*

2. *Source of infection.* Feces and eggs of domestic fowl, and feces of such household pets as dogs and cats, and of most farm animals. *Salmonella* have also been recovered in nature from numerous wild fowl, mammals and reptiles. Other sources are human cases, convalescent and passive carriers. Chronic human carriers of the animal types are relatively rare.

3. *Mode of transmission.* Epidemics are usually traced to improperly prepared foodstuffs, particularly roast fowl and dishes prepared with eggs and egg products insufficiently cooked. Little is known of the usual sources of endemic cases. They may arise by the same mechanism as epidemics or by direct or indirect transfer of infective material from the human or animal source to a susceptible individual.

4. *Prevalence.* Worldwide. Most commonly recognized in its epidemic form by the grouping of cases among individuals using a common food source. In such circumstances, a high proportion of the affected individuals develop clinical symptoms. In the endemic infections, passive cases apparently outnumber clinical cases. Detailed information on this subject is lacking.

5. *Current laboratory services.*

   A. Cultural examinations of feces for *Salmonella* organisms including serological typing service.

   B. Cultural examination of suspected food vehicles for *Salmonella* organisms.

6. *Collection of specimens.* As soon as a case or an outbreak of foodborne gastroenteritis is suspected, consult your local director of health requesting an epidemiological investigation. This is desirable for the protection of the public. Meantime make certain that no food that may have been the vehicle of infection is discarded.

   A. Use "FE" outfit. Using the sterile swab in the outfit, transfer a portion of the stool about the size of a sphere ½" in diameter to the specimen jar and emulsify it in the glycerol solution. If stools are liquid, transfer to the specimen jar with a clean spoon or other implement a quantity approximately 2 ml. in volume. Do not use spoon or other implement for more than one sampling unless carefully sterilized in the interim. Care should be taken that the stool is passed into a vessel that has been thoroughly disinfected or sterilized.

   B. Rush suspected foods to the Bureau of Laboratories for examination; do not delay. Protect suspected foods from contamination during transit to Bureau of Laboratories. If possible, refrigerate food samples during transit.

7. *Limitations of laboratory tests.* In general, the *Salmonella* types reported refer to types of animal origin. Strictly speaking, the typhoid, para-A, para-B and para-C (*hirschfeldii*) organisms, all of human origin, are also members of the *Salmonella* group. No single type of *Salmonella* is constantly associated with a definite clinical syndrome since the illness produced may range from the subclinical case to a typhoidal syndrome and especially in infants may produce septicemia, meningitis or other grave illnesses involving any tissue or organ. However, the most frequent manifestations of *Salmonella* infection, particularly with the animal strains,

are acute diarrheal conditions and acute gastroenteritis, food infections usually loosely spoken of as "food poisoning". A food handler may spread infection if he himself is infected or becomes a carrier or if he dresses meat or fowl carrying the organisms without taking proper sanitary precautions. The *Salmonella* type reported is of importance in investigating the source of an illness or outbreak.

Where laboratory findings for *Salmonella* are negative, the possibility of staphylococcus food poisoning should be considered, particularly if the incubation period of the illness appears to have been 8 hours or less. See "FOOD POISONING: BACTERIAL INTOXICATIONS".

SCABIES (THE ITCH)

1. *Etiologic agent.* *Sarcoptes scabiei,* the itch mite.

2. *Source of infestation.* Persons harboring the itch mite.

3. *Mode of transmission.* Direct contact with infected person. Indirect spread by use of towels, underclothing, gloves, bedding, etc. of such persons is less common.

4. *Prevalence.* Widespread and occuring sporadically and in epidemics, under conditions of crowding, body uncleanliness, neglect and lack of soap and water.

5. *Current laboratory services.* Identification of mites or of eggs, larvae or nymphs expressed from burrows in skin.

6. *Collection of specimens.* Place material in any convenient small vial containing 10% formalin and stopper tightly before submission to the laboratory.

7. *Limitations of laboratory tests.* Laboratory examination is seldom needed to establish or confirm a diagnosis of scabies.

SCHISTOSOMIASIS*

1. *Etiologic agent.* Three species of schistosomes mature in man, *Schistosoma mansoni, S. hematobium* and S. *japonicum.* (The larvae of certain other species of schistosomes produce schistosomiasis of birds and rodents. The larvae of these other species may penetrate the human skin and die in the skin causing a dermatitis commonly known as "swimmer's itch". The larvae of these schistosomes of lower animals do not produce schistosomiasis in humans.)

2. *Source of infection.* Persons harboring the infection and snails serving as intermediary host.

*Formerly known as Bilharziasis.

3. *Mode of transmission.* The ova of *S. hematobium* usually leave the body with the urine; those of *S. mansoni* and *S. japonicum* with the feces. The ovum hatches in water and the liberated larva or miracidium enters a suitable snail host. After growth and metamorphosis within the snail, free swimming larvae known as cercariae emerge after several weeks. These larvae penetrate the human skin, enter the blood stream and are carried to the blood vessels of the liver where they develop to maturity following which they migrate to the veins of the abdominal cavity. Those of *S. hematobium* characteristically lodge in the blood vessels of the bladder; those of *S. mansoni* and *S. japonicum* in the vessels of the rectum. The ova are deposited into the abdominal venules from which they work their way to the lumen of the bowel or bladder.

4. *Prevalence. S. mansoni* occurs in the West Indies, northeastern and eastern South America and Africa. *S. hematobium* occurs in Africa and parts of the Middle East. *S. japonicum* occurs in the Orient. None of these species is indigenous to the continental United States but *S. mansoni* is found in Puerto Rico. *S. japonicum* is endemic in the Philippines. ("Swimmer's itch", a dermal schistosome invasion, is prevalent among bathers in lakes of the North Central part of the United States.)

5. *Current laboratory services.*

   A. Microscopic examinations of feces and urine for identification of ova.

   B. Complement-fixation tests on blood specimens by arrangement with cooperating laboratories.

6. *Collection of specimens.*

   A. Collect portion of stool and urine specimens separately, placing them in bottles supplied in "PD" outfits. Mark history blank plainly "For schistosomes".

   B. Collect 5-10 ml. blood in sterile bottle in "MI" outfit. Mark history blank plainly "For schistosomiasis".

7. *Limitations of laboratory tests.*

   A. Identification of the ova is necessary to differentiate between species of *Schistosoma.* The ova of one species (*Schistosoma hemabotium)* are usually found in the urine only.

   B. Positive complement-fixation tests are indicative of infection but do not differentiate among types.

SMALLPOX (VARIOLA)

1. *Etiologic agent.* The virus of smallpox, *Borreliota variolae.*

2. *Source of infection.* Lesions of the mucous membranes and skin of infected persons.

3. *Mode of transmission.* By contact with persons sick with the disease; this contact need not be intimate, but aerial transmission except for short distances appears to be unlikely. By articles or persons freshly contaminated by discharges of the sick from lesions of his skin and mucous membranes.

4. *Prevalence.* There is no regional or climatic limitation to its prevalence except as population groups are more or less well protected by vaccination or by non-exposure to the disease. Distribution is sporadic or epidemic and varies widely according to the immunity status of the population of an area and its exposure to infection from without. Frequency is greatest in winter and least in summer.

5. *Current laboratory services.* No practicable laboratory test available at present.

STREPTOCOCCAL INFECTIONS, HEMOLYTIC

A. Scarlet fever and Streptococcal Sore Throat (Streptococcal Nasopharyngitis, Streptococcal Tonsillitis, "Septic Sore Throat")

1. *Etiologic agent.* Group A beta hemolytic streptococci of which there are at least 40 types.

2. *Source of infection.* Discharges from the nose, throat, or purulent complications of acutely ill or convalescent patients or carriers, or objects contaminated with such discharges. Nasal carriers are particularly likely to contaminate their environment.

3. *Mode of transmission.* Transmission can occur directly or indirectly. The direct route involves contact with the person of the patient, a carrier, or objects he has handled. By the indirect route streptococci are inhaled by breathing air containing these bacteria. Streptococci reach the air via contaminated floor dust, lint from bedclothing, personal clothing, handkerchiefs or occasionally in droplet nuclei discharged by coughing or sneezing. Explosive outbreaks may follow the ingestion of contaminated milk or other food.

4. *Prevalence.* Recognized cases are most common in the temperate zones, less common in the semi-tropical areas, and rare in tropical climates. Aside from food-borne epidemics, which may occur in any season, the highest incidence of scarlet fever and streptococcal sore throat in the United States occurs during the late winter and spring. Inapparent infections are as common or more common in the tropics than in the temperate zones.

5. *Current laboratory services.* Cultural examinations of swabbings from the mucosa of affected areas in the throat. Precipitin tests for grouping streptococci (Lancefield groups) are not made routinely but may be done when necessary to establish source of an outbreak. Typing of streptococci (Griffith types) can be made through arrangements with cooperating laboratories when essential.
6. *Collection of specimens.* Use "HS" outfit. Rub sterile swab provided over reddened or otherwise affected areas in the throat. Remove stopper from tube containing agar and plunge swab into jelly and leave it there. Replace stopper and return to laboratory at once.
7. *Limitations of laboratory tests.* A single negative culture should not be considered conclusive if there are clinical symptoms especially when more than 12 hours have elapsed between taking the culture and its receipt at the laboratory. In all such cases another culture should be sent.
   Positive results on throat or nose cultures are not conclusive evidence that the beta hemolytic streptococcus is the cause of the condition. These results must be considered simply as contributory to the clinical evidence. Positive results on cultures from suppurating lesions are more conclusive. Results of serologic grouping or typing have epidemiological but not diagnostic significance.

B. Erysipelas

1. *Etiologic agent.* Beta hemolytic streptococci of Group A.
2. *Source of infection.* Infected material from human sources containing hemolytic streptococci either directly from the respiratory tract of the same individual or indirectly from exogenous sources.
3. *Mode of transmission.* Bacteria enter breaks in skin either directly from same individual or indirectly from exogenous sources.
4. *Prevalence.* Geographic and seasonal distribution similar to that of streptococcal respiratory tract infections.
5. *Current laboratory services.* Cultural examinations of exudate from lesions or of pus from complicating suppurations. Precipitin tests for grouping streptococci (Lancefield groups) are not made routinely but may be done when necessary to establish sources of outbreaks. Typing of streptococci (Griffith types) when essential can be done through arrangements with cooperating laboratories.
6. *Collection of specimens.* Use "HS" outfit. Collect exudate or pus on sterile swab in outfit. Remove stopper from tube containing agar jelly and plunge swab into jelly and leave it there. Replace stopper and return to laboratory at once.
7. *Limitations of laboratory tests.* The presence of streptococci producing beta hemolysis upon blood agar is strong presumptive evidence of their causative role in suspected erysipelas. Results of serologic grouping or typing have epidemiological but not diagnostic significance.

C. Puerperal Infection (Puerperal Septicemia)

1. *Etiologic agent.* Beta hemolytic streptococci usually of Groups A, B, C or G. Infection may also be caused by anaerobic streptococci or by a mixed bacterial flora.

2. *Source of infection.* Hands and instruments used in examinations of the genital tract just prior to, during, or following confinement; the nose and throat of the parturient woman or her attendants just prior to, during, or just after confinement; infectious processes and discharges of the genital tract prior to confinement.

3. *Mode of transmission.* Direct transfer to the birth canal of infectious material by hands; transfer of bacteria discharged from the nose and throat of infected or carrier individuals. Indirectly by articles soiled by infectious discharges brought into contact with the genital tract of the patient.

4. *Prevalence.* The most common cause of preventable sickness and death related to childbearing. Epidemics occur particularly in institutions where aseptic technics are faulty.

5. *Current laboratory services.*

   A. Cultural examinations of vaginal discharges.

   B. Blood cultures.
   (Precipitin tests for serologic grouping of organisms isolated or typing of cultures through arrangement with cooperating laboratories may be done at times for epidemiological reasons.)

6. *Collection of specimens.*

   A. Use "HS" outfit. Collect vaginal discharges on sterile swab in outfit. Remove stopper from tube containing agar, plunge swab into jelly and leave it there. Replace stopper and return to laboratory at once.

   B. Use "BC" outfit. Collect 5 ml. blood aseptically with sterile syringe and needle. Introduce as directed into bottle containing nutrient medium.

7. *Limitations of laboratory tests.* The presence of streptococci producing beta hemolysis on blood agar is confirmatory of clinical evidence. Serologic grouping or typing of organisms isolated has epidemiological but not diagnostic significance.

SYPHILIS

1. *Etiologic agent.* *Treponema pallidum* (*Spirocheta pallida*).

2. *Source of infection.* Discharges from obvious or concealed lesions of the skin and mucous membranes, the semen, the blood of infected persons.

3. *Mode of transmission.* By direct contact with infected persons, chiefly by sexual intercourse, occasionally by kissing; by dental and other surgical or technical accidents, and rarely by indirect contact with articles soiled with discharges or blood containing the organism; congenitally from syphilitic mother through the placenta.

4. *Prevalence.* Widespread throughout the world varying with race, social customs, sex and age. Mostly commonly acquired between the ages of 18 and 30. Differences in racial incidence are related to social rather than biological factors.

5. *Current laboratory services.*

   A. Dark-field examinations of material from lesions for *Treponema pallidum.*

   B. Quantitative serodiagnostic tests on blood specimens.

   C. Serodiagnostic, total protein and colloidal gold tests on spinal fluids.

6. *Collection of specimens.*

   A. **For dark-field:** Use "CF" outfit. Wash lesion with sterile physiological salt solution. Rub firmly with sterile gauze. Remove all blood with sterile gauze.
   Gently compress the tissues surrounding the chancre until sufficient serum exudes to permit filling the capillary tubes. A compress of 2 per cent novacaine applied for a few minutes will aid in obtaining the deep exudate. Hold the capillary tube in a horizontal position, while the specimen is being collected. The fluid will enter the tube rapidly due to capillary attraction. Seal by pressing both ends of tube into the wax in the small vial. Replace tube in the container and mail. If any antiseptic or other local treatment has been administered, a salt solution compress should be applied and the patient instructed to return on successive days. In case an antiseptic has been used on the lesion and the regional lymph glands are enlarged, fluid from them also may be submitted. In a bilateral adenitis there may be an associated chancroidal infection and one or more of the glands may be fluctuating, inflammatory and tender. The indurated, shotty, nontender glands should be chosen. Sterilize skin over selected lymph node. Draw about 0.5 ml. of sterilized salt solution into a 1- or 2- ml. sterile syringe using a 22- or 24- gauge needle. Immobilize the gland by grasping it so that the skin is drawn tightly over it. Thrust the needle into the gland, rotate it to break apart some of

the tissue at its tip, and inject a few drops of the salt solution, taking care that the point of the needle enters the gland and not the surrounding tissues. Draw into the syringe a few drops of the fluid from the glandular tissue. The aspirated fluid (which should contain very little blood) may then be deposited from the syringe onto any glass surface, such as that of a microscope slide or the clean side of a flat bottle, and collected in capillary tubes in the same manner as described for fluid from a chancre.

B. **Blood specimens:** Use "SY" outfit. Collect about 5 ml. of Blood in the dry, sterile, chemically clean jar provided in the container. If a syringe is used it should be dry. Never use a hot syringe. Never use a syringe or a needle containing any trace of water, alcohol or oil. If syringe and needle cannot be thoroughly dried, rinse with sterile physiological salt solution. Avoid air bubbles in blood when drawing specimen; do not force blood violently from syringe into specimen vial. Replace cover or cork promptly after putting blood in vial. Always allow clot to form firmly in the vial before blood is agitated in any way. Do not expose blood to heat such as placing near a radiator or sterilizer or placing in direct sunlight; do not expose to freezing temperatures. Mail or otherwise send specimens promptly to the laboratory; avoid when possible shipping specimens over a week-end or a holiday. When specimens cannot be mailed or sent within an hour or two, place in a refrigerator but not where it will freeze.

C. **Spinal fluid:** Use "SF" outfit. Collect about 10 ml. of spinal fluid using a needle sterilized in an autoclave. Discard enough of first flow of fluid to eliminate blood present in needle which may have passed through small blood vessel. Let fluid flow directly into sterile bottle supplied with this outfit; replace cap tightly. Keep fluid in refrigerator until shipment to laboratory; do not remove blood or other material by centrifuging. Mark history blank plainly to indicate type of examination requested.

7. *Limitations of laboratory tests.*

A. **Dark-field examinations:** Positive results on specimens from the mouth should be viewed with caution since oral spirochetes of no significance have a morphology similar to *Treponema pallidum*. The report of a trained observer will always take this into consideration.
Negative results on specimens from infected individuals may result when antiseptics have been applied locally.

B. **Blood tests:** No single test for syphilis will pick up all cases which may prove positive by another standard test. Non-specific or false reactions infrequently occur in healthy individuals but may occur in febrile conditions, during the

course of upper respiratory infections, for a brief period after a successful "take" following smallpox vaccination, or in malaria and a few other diseases relatively rare in Connecticut. The physician may, however, frequently encounter the asymptomatic case of lues. For these reasons, the following suggestions are made for evaluating laboratory results obtained in Bureau of Laboratories:

a. **Suspected chancre present:** Place greatest reliance upon appearance of lesion and history of exposure. It is advisable to confirm diagnosis by dark-field examination for *Treponema pallidum.* (Use "CF" mailing outfit). Serological tests rarely show a reaction during first 3 weeks and may remain non-reactive for first 6 weeks. Usually, after 5 weeks the quantitative tests show a rapidly increasing titer in the absence of treatment when repeat specimens are examined. When treatment has been begun and completed while the serologic test for syphilis (hereafter called STS) is negative, a reaction may never be obtained.

b. **Early syphilis:** After the seronegative primary stage, untreated early syphilis, even when latent, is accompanied by strongly reactive STS. Titers rising to or above 16 dils are expected; titers 64 dils and above are not uncommon. In this state serological response to specific treatment is most pronounced. Specimens taken at suitable intervals to evaluate results of treatment should show a reduction in titer; when a decline in titer is followed by a rise of more than one dilution (e. g., from 8 dils to 32 dils), the probability is that the case will relapse clinically. If this rise in titer is confirmed by a subsequent STS, a serologic relapse has occurred and treatment is indicated. It should be emphasized that reduction in titer is not coincidental with antibiotic therapy but usually occurs during the first few months of the post-treatment period.

c. **Late syphilis including late latent:** It would be misleading to make more than a few generalizations about the behavior of STS in late syphilis. Frank reactions are anticipated when there has been no past treatment. The titer of the blood reaction may, however, not be consistent from day to day; fluctuations are common. Because many cases of late syphilis occur among persons who have received treatment that was not adequate, the titer of the reaction obtained may not prove a reliable criterion for diagnostic purposes. Any reaction by any standard test should be supplemented by careful clinical examination and the taking of a searching history. When syphilis is strongly suspected, a spinal fluid should be examined to rule out neurological involvement.

When unexpected reactions are obtained in the absence of symptoms and past treatment, the diagnosis should be held in abeyance pending consideration of repeat findings. (See below "NON-SPECIFIC REACTIONS".)

When a history of past treatment has been obtained, the physical condition of the patient and his history are more reliable guides than are STS on blood in deciding upon the need for further treatment.

Late syphilis under treatment does not follow any predictable serological pattern. Fluctuations in titer will occur even with continued physical improvement of the patient. Many cases may never become seronegative even though the disease has been arrested.

d. **Non-specific reactions:** Not all weak reactions are non-specific but non-specific reactions seldom exceed a titer of 2 dils in a well individual unless induced by vaccinia. Higher titers in the absence of syphilis may result not only from yaws, malaria, leprosy or other diseases not prevalent in Connecticut but also from febrile conditions of any sort, upper respiratory infections and recent immunization procedures. Always repeat the test when laboratory findings are suspected to have a non-specific origin. If still in doubt, repeat the test after 3 months.

e. **Negative tests:** Repeat the test if suspected primary syphilis. Reactions may not develop until 6 weeks after primary lesion occurs.

C. **Tests on spinal fluid:** A reaction to a serodiagnostic test for syphilis is considered indicative of central nervous system involvement since false reactions occur rarely or not at all with spinal fluid in a competent laboratory performing a standard test. Cell counts (preferably made locally), total protein determinations and colloidal gold tests are helpful in determining extent and type of involvement.

TETANUS (LOCKJAW)

1. *Etiologic agent.* Tetanus bacillus, *Clostridium tetani.*

2. *Source of infection.* Soil, street dust, and animal feces.

3. *Mode of transmission.* Wound infection.

4. *Prevalence.* World-wide distribution, following wound infection. Most frequent in North America among young males and in summer. Prevalent especially following wounds contaminated with manured soil. Tetanus in the newborn occurs where there is neglect of surgical asepsis and ordinary cleanliness in the care of the umbilical cord and its dressings in the first two weeks of life. The condition

is a serious factor in infant mortality where midwives are ignorant or incompetent.

5. *Current laboratory services.* Cultural examinations and animal inoculation tests of curettings from wounds.

6. *Collection of specimens.* Place tissue fragments from curetted wound in sterile bottle in "MI" outfit. Mark history blank plainly "For tetanus".

7. *Limitations of laboratory tests.* Animal inoculation tests are more reliable than cultures; a negative culture is not conclusive.

TRACHOMA

1. *Etiologic agent.* The virus of trachoma (*Chlamydozoon trachomatis),* related to the psittacosis-lymphogranuloma venereum group.

2. *Source of infection.* Secretions from the eyes and mucoid or purulent discharges of the nasal mucous membranes of infected persons; tears of such persons also carry the infections.

3. *Mode of transmission.* By direct contact with secretions of infected persons and by indirect contact with materials recently soiled with their infective secretions. Flies have been incriminated in Eastern countries.

4. *Prevalence.* World-wide, but distribution very unequal in different countries and continental areas.

5. *Current laboratory services.* None available.

TREMATODE INFECTIONS*, MISCELLANEOUS

1. *Etiologic agent.* Among others are *Fasciola hepatica* (sheep liver fluke), *Fasciolopsis buski* (giant intestinal fluke), *Clonorchis sinensis* (Chinese liver fluke), *Paragonimus westermani* (Oriental lung fluke), *Echinostoma ilocanum* (Garrison's fluke), *Heterophyes heterophyes* (minute fluke), *Metagonimus yokogawai* (Yokogawa's fluke).

2. *Source of infection.* Infected humans or other animals that may serve as a primary host.

3. *Mode of transmission.* All of the flukes have a complex life cycle and infection of the human takes place by ingestion of the infective form of the cercaria in water, on aquatic plants, in raw vegetables, fish, crayfish, etc.

4. *Prevalence.* For the most part limited to oriental regions. *Fasciola hepatica* is present in Europe and in the western hemisphere.

5. *Current laboratory services.* Microscopic examinations of feces for ova; similarly, examinations of sputum for ova of lung fluke (*Paragonimus).*

*See also "Schistosomiasis".

6. *Collection of specimens.* Use "PD" outfit. Place material in bottle furnished. Mark history blank plainly "For trematodes".

7. *Limitations of laboratory tests.* Identification of species by study of the ova is possible by an observer with special training.

TRICHINOSIS

1. *Etiologic agent. Trichinella spiralis.*

2. *Source of infection.* Uncooked or insufficiently cooked pork or pork products. Occasionally meat of fox, wolf or bear.

3. *Mode of transmission.* Only through consumption of meat, usually pork, containing viable larvae.

4. *Prevalence.* World-wide, but rare or absent in native populations of the tropics. The parasite is particularly widespread in the United States, about 1 in every 6 necropsies showing infection. Clinical cases probably occur more frequently than is indicated by morbidity reports and the disease is probably often confused with other illnesses. No selection by age, sex, race, region, season, or climate except as these affect the eating of insufficiently cooked flesh of infected hogs.

5. *Current laboratory services.*

   A. Examination of blood smears for eosinophilia.

   B. Serologic tests on blood serum (preferably during fourth week of disease) available through cooperating laboratories.

   C. Examination of laked blood and spinal fluid for larvae of *Trichinella spiralis.*

   D. Examination of biopsy tissue (usually from deltoid or pectoral muscle) obtained after 10th day of illness for encysted trichina.

   E. Examination of suspected meat or meat products for larvae of *Trichinella spiralis.*

6. *Collection of specimens.*

   A. Collect thin blood smears. Use "MA" outfit. Follow directions given under "MALARIA" for thin smears only. Mark history blank plainly "For trichinosis".

   B. Blood: Use "MI" outfit. Collect aseptically about 5 ml. of venous blood and place in sterile bottle in outfit. Allow to clot firmly before sending to laboratory. Mark history blank plainly "For trichinosis".

   C. In severe cases during the time of high fever (second or third week) collect 5 ml. of venous blood and add immediately to 15 ml. of 3% acetic acid. A suitable outfit for collection of

the blood will be furnished upon request to the Bureau of Laboratories. Mark history blank plainly "For trichina".

D. In cases which develop severe pain, stiffness and disability (usually about the 10th day of illness), the physician may in rare instances wish to excise a bit of muscle from the deltoid or pectoral muscle near its insertion for examination for encysted trichinae. Place excised tissue in sterile bottle in "MI" outfit. Mark history blank plainly "Biopsy material for trichina".

E. **Meat:** Submit a representative sample of the suspected meat in any convenient container. Mark accompanying request plainly "For trichina".

7. *Limitations of laboratory tests.*

A. **Eosinophilia:** Although not specific, marked eosinophilia lends support to clinical evidence of this disease. It occurs early enough to be a distinct diagnostic aid.

B. **Serological tests:** A reaction is seldom obtained before the third week of the disease and may not be found until the fourth or fifth week.

C. **Trichinae in blood or spinal fluid:** It is possible to demonstrate the trichinae in laked blood ordinarily only in more severe cases. A negative result is not conclusive. The spinal fluid may contain larvae when they are not demonstrable in the blood.

D. **Trichinae in biopsy tissue:** This test is of distinct diagnostic value but is not recommended except when absolutely necessary to establish a diagnosis.

E. **Trichinae in meat:** It is sometimes very difficult to establish the presence of trichinae in suspected meat, particularly ground pork products, and the examination must be carried out painstakingly. A negative result is not entirely conclusive.

Note: During the early acute gastro-enteritis adult worms may occasionally be found in feces or in vomitus. However, although present in the intestinal contents at autopsy of fatal cases, the worms are usually marred beyond recognition in body discharges during life.

(The intradermal test is a valuable diagnostic aid.)

TRICHURIS INFECTION (WHIPWORMS)

1. *Etiologic agent. Trichuris trichiura,* the whipworm.

2. *Source of infection.* Infected humans.

3. *Mode of transmission.* Ingestion with food or otherwise of embryonated ova from soil contaminated by excretions. Flies may be an intermediate factor in contaminating food.

4. *Prevalence.* Widely distributed in warm, moist regions where general sanitation is poor.

5. *Current laboratory services.* Microscopic examinations of feces for ova.

6. *Collection of specimens.* Use "PD" outfit. With a tongue depressor or similar implement take specimens of feces from different parts of the stool shortly after it is passed and place in bottles supplied with outfit.

7. *Limitations of laboratory tests. Trichuris trichiura* infection is usually asymptomatic unless very heavy. The worm may be present together with other parasites.

TRYPANOSOMIASIS, AFRICAN (SLEEPING SICKNESS)

1. *Etiologic agent. Trypanosoma gambiense* and *T. rhodesiense.*

2. *Source of infection.* Infected persons are the main reservoir in *T. gambiense* infection, while in *T. rhodesiense* infection a number of wild game, especially antelopes, act as the main reservoir. Domestic cattle and pigs may be sources of infection for the insect vector *Glossina.*

3. *Mode of transmission.* By certain species of tsetse flies, *Glossina.* There are four species of *Glossina* concerned in the transmission of sleeping sickness, *G. palpalis, G. tachinoides, G. morsitans* and *G. swynnertonii;* in nature the first two transmit *T. gambiense* infection and the latter two the *T. rhodesiense* infection, though in the laboratory many other species are capable of transmitting both infections. The fly is infected by biting an infected person or animal. The parasite develops in the body of the fly for a variable period of time, usually 18 days, the cycle depending on temperature and other factors. Infection is conveyed by the bite. The metacyclic forms are injected with the salivary gland secretion into the wound made by the tsetse fly's proboscis. Direct mechanical transmission by blood on proboscis of *Glossina* is probably responsible in some epidemics.
Once infected, a tsetse fly remains infected until it dies. The infection is not passed from generation to generation in the tsetse fly. A few cases of congenital infection in man have been reported, and transmission is also said to occur during coitus.

4. *Prevalence.* The disease is confined to tropical Africa, between 15° N. and 20° S. The distribution corresponds with that of the tsetse fly.

   T. gambiense: Gambia, Liberia, Sierra Leone, Gold Coast, Congo, S. Sudan, Uganda, Ngamiland.

   T. rhodesiense: A much more limited distribution. The southeast corner of tropical Africa, north and, in few areas south, of the Zambesi River, Mozambique, Nyasaland, Rhodesia, and Tanganyika.

5. *Current laboratory services.*

   A. **In early stages:** Microscopic examination of blood or material aspirated from enlarged lymph glands; animal inoculation tests on same material.

   B. **In later stages:** Similar examinations of cerebrospinal fluid.

6. *Collection of specimens*:

   A. **Blood films:** Use "MA" outfit. Make thick and thin films as directed. Mark history blank plainly "For trypanosomiasis".

   B. Whole blood: Use "MI" outfit. Collect 5-10 ml. of blood. Mark history blank plainly "For trypanosomiasis".

   C. **Cerebrospinal fluid:** Use "SF" or "MI" outfit. Collect 5-10 ml. of spinal fluid. Mark history blank plainly "For trypanosomiasis".

7. *Limitations of laboratory tests.* The presence of the trypanosomes is confirmatory of clinical suspicion. Only an experienced observer should attempt these examinations.

TRYPANOSOMIASIS, AMERICAN (CHAGAS' DISEASE)

1. *Etiologic agent. Trypanosoma cruzi.*

2. *Source of infection.* Infected persons and a number of domestic and wild animals, such as dogs, cats, opossums, and armadillos.

3. *Mode of transmission.* By the fecal material of infected insect vectors, various blood sucking species of *Reduviidae* (conenosed bugs), especially the genus *Triatoma,* which frequently attack man. Contamination with infected fecal material from the bug, of the conjunctivae, mucous membranes, abrasions, or wounds in the skin made by the bite of the insect. It is probably not transmitted by the actual act of biting.

4. *Prevalence.* The disease has a wide geographic distribution in Central and South America. Cases have been found in Southern Mexico. No human case has been reported as yet in the United States but several species of the genus *Triatoma* have been shown

to be carriers of *Trypanosoma cruzi* in Texas, New Mexico, Arizona, and California, and wild rodents, armadillos and opossums have been found infected in these areas.

5. *Current laboratory services.*

   A. Complement fixation tests on blood through cooperating laboratories.

   B. Microscopic examinations of thick blood films.

   C. Animal inoculation tests and microscopic examination of citrated blood.

6. *Collection of specimens.*

   A. Collect 5-10 ml. of blood in sterile bottle in "MI" outfit. Mark history blank plainly "For complement fixation test for trypanosomiasis".

   B. Collect thick blood films as for malaria (See "MALARIA"). Use "MA" outfit. Mark history blank plainly "For trypanosomes".

   C. Collect citrated blood for microscopic examination and for animal inoculation as follows: Withdraw 9 ml. venous blood and add it immediately to tube or bottle containing 1 ml. of 6% sodium citrate solution. Mix well but gently. A special outfit for this purpose will be furnished by the Bureau of Laboratories upon request. Mark history blank plainly "For trypanosomes".

7. *Limitations of laboratory tests.*

   A. The complement fixation test is considered the best single diagnostic aid.

   B and C. These procedures are successful in only a small percentage of cases. Negative findings are of no significance.

TSUTSUGAMUSHI DISEASE (SCRUB TYPHUS)*, MITE-BORNE

1. *Etiologic agent. Rickettsia tsutsugamushi (R. orientalis).*

2. *Source of infection.* Infected larval mites of *Trombicula akamushi* and related species varying with locality. The nymph and adult are free-living. The infection is passed from generation to generation and maintained by feeding upon susceptible wild rodents, particularly mice and rats of different species, varying with locality.

3. *Mode of transmission.* By the bite of infected mites.

4. *Prevalence.* Limited localities in countries of southeastern Asia, particularly India, Burma, Federated Malay States and French

*Also known as "Keldani fever", "Japanese river fever", "flood fever" and probably is identical with diseases called "tropical typhus", "rural typhus", "mite fever" and "Indian tick typhus".

Indo-China; in the island archipelagoes of the west and south Pacific, Japan, Formosa, Sumatra, Java, New Guinea, and in North Queensland, Australia. Near the Equator transmission may occur throughout the year; in Japan it is limited to the summer months.

5. *Current laboratory services.*

   A. Complement-fixation tests for rickettsial diseases.

   B. Agglutination tests (Weil-Felix) on blood specimens.

6. *Collection of specimens.*

   A. **For complement-fixation tests:** Use "MI" outfit. Collect 10-20 ml. of venous blood during acute phase and repeat about 3 weeks later. Mark history blank plainly "For rickettsial diseases".

   B. **For agglutination tests:** Collect aseptically about 5 ml. of venous blood 9 or 10 days after onset and place in sterile bottle in "TY" or "MI" outfit. Allow to clot firmly before sending to laboratory. Mark history blank plainly "For OXK agglutination". Collect another specimen during the third week of illness.

7. *Limitations of laboratory tests.* Positive complement-fixation tests are confirmatory of clinical findings and usually indicate the type of infection present. The reaction is usually negative until the 9th or 10th day after onset. In this disease complement-fixation tests serve the purpose of aiding in the elimination of typhus and Rocky Mountain spotted fevers.

   Weak or moderate results by the Weil-Felix test (OXK antigen) are of little diagnostic significance unless confirmed by a sharply rising titer during the third week of the disease. Negative results may be obtained as late as the 14th day of illness.

TUBERCULOSIS, PULMONARY

1. *Etiologic agent.* Tubercle bacillus (human, *Mycobacterium tuberculosis v. hominis);* bovine type has been found in occasional cases in some areas (outside the continental United States) where milk is not pasteurized and infection of cattle is prevalent; avian type rare and a doubtful cause of human infections.

2. *Source of infection.* Persons with "open" pulmonary tuberculosis; rarely tuberculous cattle.

3. *Mode of transmission.* Usually through the discharges of the respiratory tract, by direct or indirect contact with infected persons, by means of coughing, sneezing, or other droplet infection, by kissing; less certainly by the use of contaminated eating and drinking utensils, or by contaminated flies and dust. Infection rarely occurs from casual contact, but usually results from the continued and intimate exposure characteristic of family relationships.

4. *Prevalence.* Among the most common communicable diseases of man, with less variation in prevalence of infection according to race than in mortality. In most occidental nations its incidence and mortality are declining. Age at which first infection occurs varies; children exposed in the household and in cities are infected earlier than rural children and those not so exposed, who may escape infection until adolescence or adult age. Mortality high among infants, among adult males up to old age, and among adolescent and young adult females. An important cause of death at ages 15 to 34. Aboriginal races when first exposed develop the disease in a rapidly fatal form. Under favorable conditions for the disease it may occur at times in epidemics.

5. *Current laboratory services.* Microscopic and cultural examinations of sputum for *Mycobacterium tuberculosis.*

6. *Collection of specimens.* Use "TC" outfit. Purulent, cheesy or mucopurulent sputum, preferably that coughed up from the lungs early in the morning, is much more likely to contain tubercle bacilli than just mouth saliva. The presence of saliva, mucous, blood or stomach contents in the specimen is undesirable for reliable examination.

7. *Limitations of laboratory tests.* Failure to find *Mycobacterium tuberculosis* microscopically, even with concentration methods, does not exclude pulmonary tuberculosis. Positive microscopic findings occasionally may be due to the presence of acid-fast bacilli other than those of tuberculosis. Cultural confirmation of results is desirable in either case.

   Negative cultural findings are of no value if preservative has been added to the original specimen. In the absence of preservative, negative cultural findings on specimens which are positive microscopically are unusual and a repeat specimen should be submitted; when acid-fast organisms other than tubercle bacilli are isolated from such specimens, a special report will be sent. A negative microscopical or cultural result does not exclude the possibility of tuberculosis infection.

   Positive cultural findings on specimens which were negative microscopically do not constitute a conflicting report since the cultural method is a more sensitive means of detecting small numbers of tubercle bacilli than is the microscopic examination although it requires a much longer time for completion. Animal inoculation tests of cultures isolated are sometimes desirable to demonstrate that the culture of acid-fast organisms isolated has been found to possess the virulence characteristic of *Mycobacterium tuberculosis.*

TUBERCULOSIS, OTHER THAN PULMONARY

1. *Etiologic agent.* Tubercle bacillus (human and bovine), *Mycobacterium tuberculosis (hominis et bovis).*

2. *Source of infection.* Persons with "open" pulmonary tuberculosis, less frequently tuberculous cattle, raw milk from tuberculous cattle.

3. *Mode of transmission.* By direct contact with infected persons, by contaminated food, mainly unpasteurized milk, and possibly by contact with articles freshly soiled with the discharges of infected persons.

4. *Prevalence.* Much less common than the pulmonary form and more rapidly falling in incidence, representing in the United States less than 10 per cent of total cases and deaths from the disease. In England and Wales, the corresponding figures are 19 per cent of total notifications of tuberculosis and 16 per cent of deaths from all forms of tuberculosis. Especially common in infants and young children where intimately exposed to parental infection and to bovine infection through raw milk from tuberculous cattle.

5. *Current laboratory services.* Microscopic examination, cultures, and animal inoculation tests on pus, exudates, and body fluids for presence of *Mycobacterium tuberculosis.*

6. *Collection of specimens.* Collect material aseptically and place in sterile bottle in "MI" outfit. Mark history blank plainly "For tuberculosis" and state type of specimen. Use "SF" outfit for collection of spinal fluid in cases of suspected tuberculous meningitis.

7. *Limitations of laboratory tests.* Positive microscopic results are suggestive of infection; positive cultures and animal inoculation tests are conclusive. Negative microscopic findings may be of little value but negative cultures and animal inoculations are reliable in most instances when specimens have been properly collected.

TULAREMIA

1. *Etiologic agent. Pasteurella tularensis (Bacterium tularense).*

2. *Source of infection.* Wild rabbits and hares, deer fly (*Chrysops discalis),* wood tick (*Dermacentor andersoni* and *Dermacentor variabilis),* woodchuck, coyote, muskrat, opossum, tree squirrel, quail, skunk, water rat of Europe (*Arvicola amphibius),* cat, deer, dog, fox, hog, sage hen, and bull snake.

3. *Mode of transmission.* By bites of infected flies and ticks and by inoculation of skin or conjunctival sac, through handling infected animals, as in skinning, dressing, or performing necropsies on infected animals, or by fluids from infected flies, ticks, rabbits, and woodchucks. Ingestion of insufficiently cooked rabbit meat. Rare cases occur from bites of coyotes, skunks, hogs, cats, and dogs, where the mouth of the animal was presumably contaminated from eating

infected rabbits. Drinking contaminated water. Laboratory infections are not infrequent.

4. *Prevalence.* The disease occurs throughout North America, and in many parts of continental Europe and in Japan. In the United States it occurs in every month of the year, but especially during the rabbit hunting season. The case fatality is about 5 per cent.

5. *Current laboratory services.*

   A. Agglutination tests on blood specimens.

   B. Bacteriological examinations of material from lesions and bacteriological and pathological examination of animals or organs from animals suspected of transmitting tularemia are not ordinarily recommended but can be arranged under unusual conditions through cooperating laboratories.

6. *Collection of specimens.*

   A. After the 10th day of illness, collect aseptically 5-10 ml. of venous blood in the sterile bottle in "TY" outfit; mark history blank plainly "For tularemia". Follow up with specimen taken during third week of illness.

   B. Do not collect any other type of specimen until you have contacted Bureau of Laboratories for special instructions.

7. *Limitations of laboratory tests.*

   A. **Agglutination tests:** The agglutination test for tularemia does not become positive until the 10th day of illness and reaches its maximum reaction 14-18 days after onset. Serial specimens to detect rising titers are often helpful. The test remains positive indefinitely following infection; hence, previous history is important. False reactions are known to occur only in brucellosis.

   B. **Bacteriological studies:** Demonstration of *Pasteurella tularensis* in material from lesions or in suspected vectors is a difficult laboratory procedure involving extreme danger to laboratory workers. It is not a recommended procedure. A trained animal pathologist can recognize the lesions of tularemia in rodents by gross study at autopsy.

TYPHOID FEVER

1. *Etiologic agent.* Typhoid bacillus, *Salmonella typhosa (Eberthella typhosa).* Several types are readily identifiable on the basis of bacteriophage susceptibility.

2. *Source of infection.* Feces and urine of infected individuals and carriers. About 2 to 5 per cent of patients become permanent carriers. Family contacts may be transient carriers.

3. *Mode of transmission.* Transfer of typhoid bacilli in feces through direct or indirect contact with patient or carrier. Principal vehicles for indirect spread are contaminated food, water, milk, and shellfish and, under some conditions, flies.

4. *Prevalence.* Widespread throughout the world. Formerly endemic and epidemic in most large cities of North America and in many rural areas. Still endemic in some rural areas of the United States, but commonly now occurring as sporadic cases and as small contact and carrier epidemics. Steadily falling in incidence, particularly in urban areas supplied with safe water and pasteurized milk, and where human feces are disposed of without contaminating water supplies, food, milk, or surface of the soil.

5. *Current laboratory services.*

   A. Blood cultures (1-7 days' duration of disease).

   B. Agglutination tests (after 10th day of illness). Cultures are made from the clots after the serum has been removed.

   C. Feces, urine and bile cultures for isolation and typing of organism.

   D. Tests to detect Vi agglutinins in blood of suspected carriers.

   E. Bacteriophage typing of typhoid cultures for epidemiological purposes (made routinely on all cultures isolated at Bureau of Laboratories).

6. *Collection of specimens.*

   A. **Blood cultures:** Use "BC" outfit. Collect aseptically venous blood for culture in sterile syringe of 5-10 ml. capacity. Preferably replace needle used with another sterile unused one. Plunge needle through thin rubber top of stopper (previously swabbed with alcohol) and expel contents of syringe into medium in bottle. Rush to laboratory.

   B. **Agglutination tests and clot cultures:** Collect 5-10 ml. venous blood aseptically in sterile bottle in "TY" outfit. Allow to clot firmly before sending to laboratory. Follow early specimens by specimens taken during the third week of illness.

   C. **Feces:** Use "FE" outfit. Using the sterile swab in the outfit, transfer a portion of the stool about the size of a sphere ½ inch in diameter to the specimen jar and emulsify it in glycerol solution. If stools are liquid, transfer to the specimen jar with a clean spoon or other implement a quantity approximately 2 ml. in volume. Do not use spoon or other implement for more than one sampling unless carefully sterilized in the interim. Care should be taken that the stool is passed into a vessel that has been thoroughly disinfected or sterilized.

   Urine: Use "UR" outfit. Have patient void urine into previously sterilized vessel and place 10-15 ml. in sterile bottle provided in outfit.

Bile: Collect this material ordinarily only when carrier has been hospitalized for release from carrier state under the provisions of the Sanitary Code of Connecticut. Use duodenal tube and place 10-15 ml. of biliary drainage in sterile bottle in "FE" outfit. Be sure to check history blank to indicate specimen is bile.

D. **Blood for Vi agglutinins:** Follow directions under "B" above; mark history blank plainly "For Vi agglutinins".

E. **Bacteriophage typing:** Heads of local laboratories may submit cultures for this service.

7. *Limitations of laboratory tests.*

A. For early diagnosis a blood culture is the best diagnostic aid. It should be taken during the first five days of illness, after which the organisms usually disappear from the blood stream. Isolation of a serologically typed *Salmonella typhosa* establishes the cause of illness.

B. Agglutinins do not appear in the blood until 10-14 days after onset. The titer reaches its height during the third week of illness. Agglutinins produced as a result of previous infection or of prophylactic vaccination may persist for an indefinite period of time. Typhoid "O" agglutinins resulting from immunization do not ordinarily persist longer than 3-6 months after inoculation but typhoid "H" agglutinins may persist longer. Hence, a high typhoid "O" titer is of considerable significance when clinical findings are suggestive. Such findings must, however, be confirmed by positive blood or feces cultures. A rising titer in serial specimens is, however, highly suggestive of infection.

Results of clot cultures have the same significance as those on blood cultures (See A. above). Negative results are, however, likely to occur after the first five days of illness.

C. Isolation of serologically typed *Salmonella typhosa* from the feces, urine or bile may mean the patient is a temporary or chronic carrier or that the organism isolated is the cause of the illness. Hence, the significance of positive results must be considered in the light of clinical findings. For release of cases and of carriers, see the requirements of the Sanitary Code of Connecticut.

D. Absence of Vi agglutinins is not conclusive evidence of absence of carrier state. Presence of Vi agglutinins simply directs attention to possible carriers for cultural confirmation. The test is of most value for rapid screening purposes among large numbers of suspects.

E. It is well established that typhoid strains do not vary with respect to bacteriophage type in establishing relationship between cases and carriers.

TYPHUS FEVER

A. Epidemic or Classical Typhus (Louse-Borne) (Brill's Disease)

1. *Etiologic agent. Rickettsia prowazekii.*
2. *Source of infection.* Infected persons.
3. *Mode of transmission.* The infectious agent is transmitted by lice (*Pediculus humanus)* which have fed upon infected persons. The rickettsias are inoculated by crushing the infected lice or scratching louse feces into the wound made by the bite or into other superficial skin abrasions. Louse feces from dirty clothing and transmitted through the air to the respiratory tract may be the source of infection.
4. *Prevalence.* Widely distributed among people living under crowded and unhygienic conditions. Cases occur throughout the year with seasonal increase during the colder months.
5. *Current laboratory services.* Complement-fixation tests for rickettsial diseases.
6. *Collection of specimens.* Use "MI" outfit. Collect 10-20 ml. of venous blood during the acute phase and repeat about 3 weeks later. Mark history blank plainly "For rickettsial diseases".
7. *Limitations of laboratory tests.* Positive complement-fixation tests are confirmatory of clinical findings and usually indicate the type of infection present. The reaction is usually negative until the 9th or 10th day after onset; it is preferable to show a rising titer by comparing results of the two specimens recommended.

B. Endemic or Murine Typhus (Flea-Borne)

1. *Etiologic agent. Rickettsia typhi (Rickettsia mooseri).*
2. *Source of infection.* Infected rodents, especially *Rattus rattus* and *Rattus norvegicus.*
3. *Mode of transmission.* The agent is transmitted from rodent to man by a flea, commonly *Xenopsylla cheopis.*
4. *Prevalence.* Widely distributed in temperate, semi-tropical and tropical areas. Transmission to man occurs throughout the year, with seasonal increase during the warmer months.
5. *Current laboratory services.* Complement-fixation tests for rickettsial diseases.
6. *Collection of specimens.* Use "MI" outfit. Collect 10-20 ml. of venous blood during the acute phase and repeat about 3 weeks later. Mark history blank plainly "For rickettsial diseases."
7. *Limitations of laboratory tests.* Positive complement-fixation tests are confirmatory of clinical findings and usually indicate the type

of infection present. The reaction is usually negative until the 9th or 10th day after onset; preferably a rise in titer between the acute phase specimen and the specimen taken 3-4 weeks later should be shown.

VINCENT'S INFECTION (TRENCH MOUTH), FUSOSPIROCHETOSIS

1. *Etiologic agent.* Complex; fusospirochetal flora (fusiform bacteria and *Borrelia vincentii)* proliferates rapidly under pathologic conditions.
2. *Source of infection.* Discharges from the lesions of infected persons or from carriers.
3. *Mode of transmission.* Direct contact with infected persons or carriers and probably with articles freshly soiled by such persons.
4. *Prevalence.* Sporadic in general population. More common among persons of low nutrition and neglected hygiene. More common in children and young adults, and often occurs in outbreak form.
5. *Current laboratory services.* Microscopic examination of smears of exudate or pus from affected areas.
6. *Collection of specimens.* Use "VI" outfit. With sterile swab provided collect exudate or pus from affected areas and make smears on microscope slides provided. The use of a wire loop for the collection of material for examination is sometimes convenient.
7. *Limitations of laboratory tests.* The organisms usually associated with Vincent's infection are sometimes found on apparently healthy mucous membranes, particularly those of the mouth, in the absence of lesions. For that reason a positive result should be interpreted with caution, placing the most reliance upon the clinical picture. Since these organisms grow readily in diseased tissues, their presence is not proof of a primary etiological role.

VULVOVAGINITIS IN CHILDREN

1. *Etiologic agent.* A variety of organisms, including *Neisseria gonorrheae* (the gonococcus).
2. *Source of infection.* Discharges of infected persons.
3. *Mode of transmission.* By direct contact with infected persons and by contact with articles freshly soiled with the discharges of such persons.
4. *Prevalence.* Widespread; most common in families where there are overcrowding, neglect in personal cleanliness, and ignorance as to sanitary precautions. Epidemics are observed most frequently in institutions for children, day nurseries and schools.

5. *Current laboratory services.* Microscopic and cultural examinations of discharges for causative organism.

6. *Collection of specimens.*

   A. Smears: Use "GC" outfit. Collect vaginal discharges on swab provided and make thin smears on microscope slides. Allow smears to dry before shipment to laboratory.

   B. Cultures: Use "HS" outfit. Collect vaginal discharges on sterile swab provided, then plunge swab into agar jelly in tube furnished. Stopper and send to laboratory. Mark history blank plainly "For identification of organism".

7. *Limitations of laboratory tests.* Gonococcal infections are marked by the presence of gram-negative intracellular diplococci observed microscopically. Since the gonococcus does not withstand transportation through the mails, cultures cannot be made in a central laboratory serving the entire state. Presence of other pathogenic bacteria in cultures is presumptive evidence of their etiological significance.

YAWS (FRAMBESIA)

1. *Etiologic agent. Treponema pertenue.*

2. *Source of infection.* Discharges from skin lesions and mucous membranes.

3. *Mode of transmission.* Direct contact with lesions of patient and by non-biting flies which convey the discharges of infected persons to others.

4. *Prevalence.* At present not known to be indigenous in continental North America. Especially prevalent in the Caribbean area: (Jamaica, Haiti, Trinidad, Antigua, and other islands of the Leeward group), and more in some villages than others; also in some coastal and valley settlements of Colombia.

5. *Current laboratory services.* See "SYPHILIS".

6. *Collection of specimens.* See "SYPHILIS".

7. *Limitations of laboratory tests.* There is no reliable way of distinguishing between the *Treponema pertenue* of yaws and the *Treponema pallidum* of syphilis or between the serological reactions evoked by both.

YELLOW FEVER

1. *Etiologic agent.* The virus of yellow fever, *Charon evagatus.*

2. *Source of infection.* The blood of infected persons, monkeys, marmosets.

3. *Mode of transmission.* In cities and on board vessels by the bite of infected *Aedes aegypti* mosquito. In the forests of South America by *Hemagogus spegazzini* and some other mosquitoes. In Africa rural and forest transmission occurs through the bite of *Aedes simpsoni, Aedes africanus* and other mosquitoes. (There is no evidence to incriminate orthropods other than mosquitoes).

4. *Prevalence.* Not known in the Pacific Basin. No cases have occurred in West Indies, Mexico or the United States since 1925. Endemic among human beings and some animals. Epizootic among primates (and perhaps other jungle animals) of all countries of South America except Uruguay and Chile. It is still endemic in the jungle form in Panama. Occasional human cases and even epidemics occur in Africa and South America.

5. *Current laboratory services.* None available at present at Bureau of Laboratories. Special serological tests and other virus studies are done at a few virus laboratories throughout the country.

6. *Collection of specimens.* Contact Bureau of Laboratories to determine if studies at a cooperating laboratory can be arranged; instructions will be given at that time.

7. *Limitations of laboratory tests.* Lack of facilities constitutes the main limitation at present. Demonstration of increasing titer of neutralizing antibodies during course of disease is of diagnostic value.

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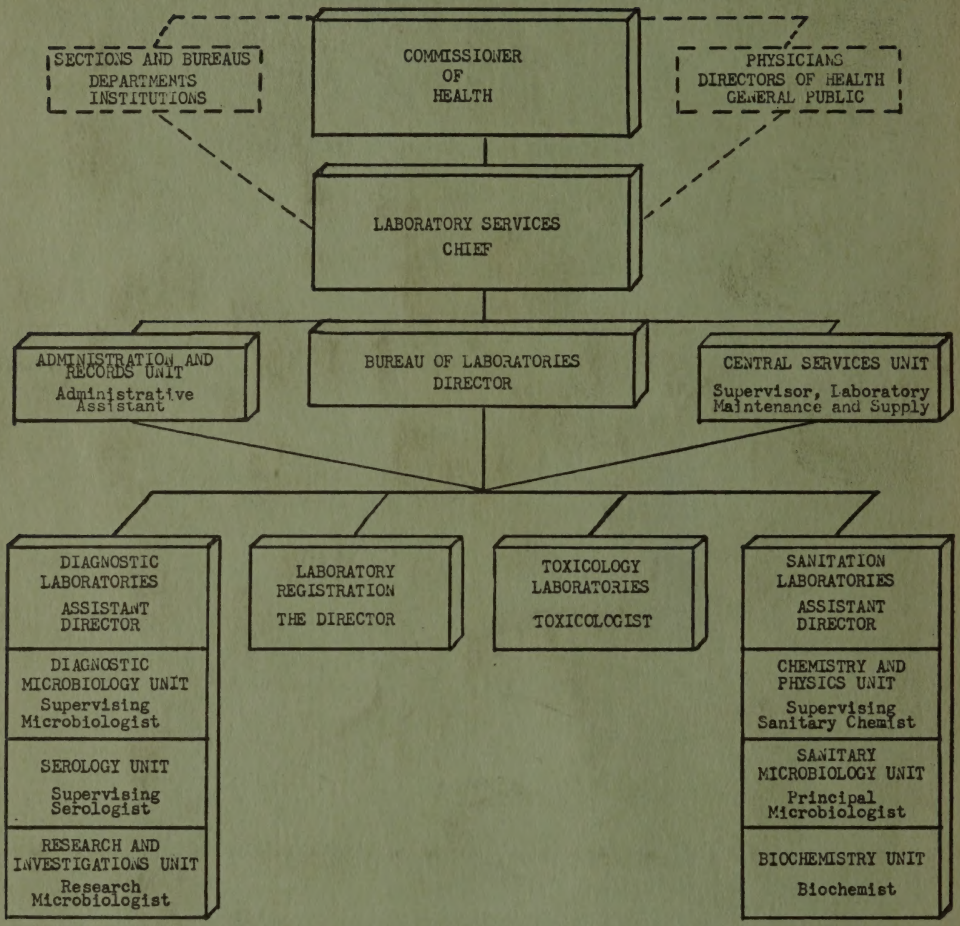

Connecticut State Department of Health

Stanley H. Osborn, M. D., Dr. P. H., Commissioner

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